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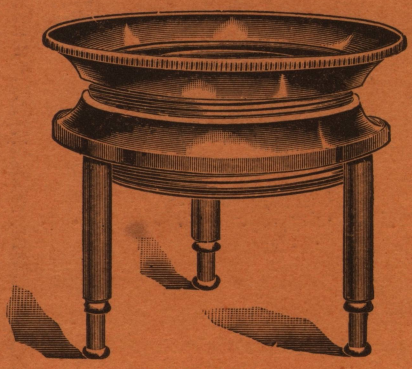

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The Value of Methylen Blue as an Intravitam Stain in the Tunicata.

While working for special results on the tunicate nervous system, with methylen blue, I found that this anilin could be made of much value as a general stain where living material was obtainable. Small species, such as *Amorœcium*, *Botryllus*, and *Perophora*, as well as young *Molgulæ*, left in sea water containing just enough methylen blue to color the water a lively blue (about 1 part to 5000) for half an hour, will give almost diagrammatically the branchial basket and its organs, as well as the free mesenchyme cells of the body cavity, leucocytes and phagocytes. This method is especially favorable for cilia; the demonstration of cilia in motion, the arrangement of cilia in rows on the surface of the cell, and the peculiar thickened basal portion of the tunicate cilium can all be well shown. Long, whip-like flagellæ, which are found in the endostyle and ciliated funnel, also take the blue and stand out with wonderful distinctness. The above named cells are usually the first to stain. The sensory or peripheral portion of the nervous system stains relatively early (from 1 to 1½ hours after immersion), while the deeper lying nerve cells and motor fibers stain later. Cells of the central nervous system are sometimes found colored blue as much as five hours after immersion. But all of this so-called staining of the different tissues is transitory, sometimes lasting only a few minutes—as in the case of the very delicate neurofibrils—or for several hours, or even days, in the case of the mesenchyme cells.

Special Methods.—The best results for the staining of the nervous system were obtained by the two following methods: Molgulæ were placed in a weak solution (1–5000) of Meyer's BX methylen blue in sea water, and allowed to remain from one to five hours, according to the size of the animal and the tissue to be stained. For staining by immersion small specimens were used. It was found necessary to have the animals absolutely fresh, or satisfactory results could not be depended upon. Molgulæ which had remained in aquaria for so short a period as two to three days, frequently refuse to take the stain; or give a diffuse staining. The exact intensity of the blue in solution does not seem to be as important a factor as the length of immersion. Just before taking out the animals

for examination they were removed to a dish of running sea water ærated by a pipette nozzle. Specimens thus treated gave uniformly good results, while those in which this process was omitted did not. Whether the former result was due to the revival of the animal or the oxygenation of the tissues, is hard to say. But the physical condition of the animal seems to play an important part.

The other method successfully used was to inject a fairly strong solution (1 to 4 per cent.) of methylen blue into the ovarian vein of Molgulæ. From thence the fluid reached the heart and was pumped to all parts of the body. A small amount of fluid should be used and great care taken not to allow too much of the body fluid to escape through the hole made by the needle. The percentage of successful staining by this method is small; but the results are valuable, because of the use of large specimens. The peripheral system seems to come out especially well by this method. Animals which die as a result of the injecting process give a diffuse staining of tissues that is of no neurological value. This diffuse staining can be recognized at once, by the fact that the whole animal stains a pale greenish blue color; while in a perfectly successful stain the color is deep blue.

The use of ammonia is advocated by Apathy in bringing out the stain. He believes that in exposure to the air just before examination, the specimen takes up some ammonia from the atmosphere. Molgulæ exposed to the fumes of ammonia gave negative results. The same may be said regarding the exposure of stained tissue to the air. It could not be proven that oxygen was a factor in the bringing out of the stain.

The fixation of the stain for permanent preservation.—Most of the methods used by investigators (*vide* Arnstein, Apathy, Bethe, Dogeil, Huber, Meyer, Peabody, Retzius, and others) agree in the use of picrate ammonia as a fixing agent. Aqueous solutions are usually employed, and the material is allowed to remain in a saturated solution of ammonium picrate from a few minutes to several hours, or even days, according to the size and permeability of the material. The macerating effect of the fluid is avoided in some cases by the addition of one part to 100 of a 1 per cent. osmic acid solution. Such material may then be either mounted permanently in a solution of saturated ammonium picrate and glycerin in equal parts (*vide* Meyer, Retzius); or in chemically pure glycerin without washing out (*vide* Dogeil); or by the somewhat complicated method of Apathy, when, after fixation in ammonium picrate plus a little (5 drops to 100 c. c.) concentrated ammonia, the material is passed through glycerin; glycerin and gum arabic; and finally mounted in a solution of gum arabic, cane sugar, and water, in equal quantities. I may say, in passing, that with tunicates I found the last named method the least useful.

By far the best of my results were obtained by the following modification of the methods of Bethe and Apathy:

Cut out the part to be used, and after examination under a fairly high power of the microscope, remove the successfully stained material to a saturated solution of ammonium picrate in sea water. The pieces of tissue are then immediately removed to a slide, or small dish, where they are left for a few minutes (10 to 20, according to the size of piece), in the following solution:

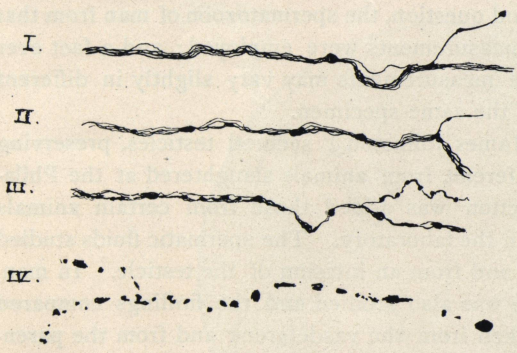
Sea water (or normal salt),	50 c. c.
Conc. c. p. glycerin,	50 c. c.
Conc. ammonia,	1 drop.

Add more glycerin, and finally mount in glycerin containing just enough of the ammonium picrate to color it slightly. Specimens put up in this way have kept their color now for over three years, no noticeable change having taken place during that time.

The above method was frequently modified. I did not find, as some later writers have done, that better results were obtained by a bath of longer duration in ammonium picrate. A short bath of from ½ to 1 minute suffices for sensory nerves and peripheral sense organs, and somewhat longer for deeper lying nerves.

I am inclined to take Apathy's view regarding the successful staining of the nerve fibers. He believes that he gets a true stain and not an impregnation. In a few very successful cases I have succeeded in following to the nerve cell a bundle of fibers, which I believe to correspond to primitive neurofibrils.

More frequently, however, an impregnation probably takes place, the whole interfibrillar space taking the blue. This can best be shown by following the successive changes which take place in the tunicate nerve fiber after death, or during the diffuse staining which takes place in the later stages of every successful nerve staining with methylen blue. The successful stain of a fiber shows an almost unbroken wavy blue line, or series of interlacing fibrils, around which can be seen very faintly the sheath. This fiber has almost no knobs or granulations in its course except at the true ending. At a point of branching a triangular blue area, probably caused by the stretching of the sheath, can be seen. As the tissue dies, however, a change takes place. The nerve fiber begins to bead, at first hardly noticably, but later these beadings become so large as to distort the whole fiber and completely change its appearance. Ultimately all trace of the fiber disappears except a line of irregularly placed blue globules. (See figure.)



Four stages in the degeneration of a nerve fibre:
I. A successful impregnation.
II. Ten minutes later; beading commencing.
III. Twenty minutes later; much beading.
IV. Two hours later.

This same beading of the fiber is frequently induced by fixation with ammonium picrate, and it is only in rare cases when we have the fiber fixed so that it shows the individual fibrils. Frequently there appears to be a vacuolization of the nerve. Large vacuoles appear, at one side of which there is a heavier deposit of blue. This is, perhaps, a pathological condition induced by the injection of the methylen blue.

Fixation for imbedding and sectioning.—Parker's sublimate and alcohol method was tried with no success. Bethe's ammonium molybdate method (Arch. f. Mik. Anat. xliv. '94) yielded poor results. His later method (Anat. Anz. xii. '96)

proved of more value. After staining tissues in a concentrated solution of ammonium picrate (which I used in sea water) the material is brought into the following solution :

Ammonium molybdate, - -	1 gr.	
H_2O, - - - -	10 gr.	} or 20 c.c. H_2O.
5 per cent. osmic, - - -	10 gr.	
Peroxide of hydrogen, - -	1 gr.	

or (with somewhat better results for tunicates), phosphomolybdate of soda may be substituted in the above formula for the ammonium molybdate.

After ¾ to 1 hour in above solution (or 4 to 12 hours in the osmic solution), we wash in water, rapidly pass through the alcohol, xylol, and imbed in paraffin. Results from this method have not been uniformly successful.

De Witt Clinton High School, N. Y. C. GEORGE WILLIAM HUNTER, JR.

Spermatozoa of Man, Domestic Animals, and Rodents.

The male cell, or spermatozoön, is of minute size, and in its locomotor energy and vitality resembles a flagellate monad. Anatomically it is a true cell, consisting of the "head," composed mainly of nucleus, and the motile "tail," which may be fibrillated, and a small central portion between the head and tail, which is sometimes regarded as the "centrosome."

In studying the spermatozoön of the mastiff, I noticed the striking resemblance it bore to that of man ; finding that if the spermatic fluids were allowed to stand for a time, even staining did not furnish sufficiently satisfactory evidence to enable one to distinguish, beyond question, the spermatozoön of man from that of the dog, except where careful measurements were employed ; and a fact ever to be borne in mind is, that these measurements may vary slightly in different persons and animals, and even in the same specimen.

Through the courtesy of Dr. James Johnston I secured testicles, preserving as much as possible of the vasdeferens, from animals slaughtered at the Philadelphia abattoir, to which collection was added those from certain animals employed for experimental work in the laboratory. The spermatic fluids studied were taken from the vasdeferens, and from an incision of the testicle. In man, the fluid ejaculated at intercourse was also studied and the findings compared with those where the fluid was taken from the vasdeferens and from the parenchyma of the testicle.

Examinations were made immediately after the testes had been removed, and on the first, second, third, fourth, and fifth days after their removal. Spermatic fluids thus collected were placed in cold water, after which it was found that the tails became coiled, and were soon detached from the heads. In no case were the spermatozoa found to possess individual movement twenty-four hours after the testicle had been removed, or after the death of the animal ; nor were they ever found motile in man even a few hours after death. These findings differ from the statements often made, that spermatozoa remain active for a long time after the death of the animal. To determine this point one testicle was kept in a cool room, and the other at a temperature of about 75° Fahr., when certain

other changes were also observed. After twenty-four hours it was common to find several free heads and tails, their number increasing daily, until by the fourth day it was often difficult to find a perfect spermatozoön; yet these, when present, showed evidence of marked degeneration, and their reaction to staining was not constant.

Staining was accomplished by the various anilin dyes, of which carbol-fuchsin was found to be of most value, therefore the accompanying illustrations were sketched from specimen slides stained by carbol-fuchsin. In examining specimens stained in 1899, I find that the tail is the first to give up its stain, and from one-fourth to one-seventh of the tails of the spermatozoa of the sheep and rats show no stain, and are seen with difficulty. Fading was noted to take place earlier where methylen blue was employed. In but one instance, that of the mouse, was it necessary to apply heat in order to stain the spermatozoa.

Measurements were made by the use of both the stage and eye-piece micrometers, always measuring the entire length, dimensions of head, and length of tail; the latter being markedly altered whenever the staining was imperfect. It was thought to be of possible service to have all measurements recorded both in millimeters and in inches. All measurements and sketches were made with a 1-6 objective and 2 eye-piece. In the sketching no attempt was made to preserve the original size of the cells. The total lengths given were obtained by the measuring of complete cells, recording the greatest and smallest measurements only; while the measurements of the heads and tails were often taken after these parts had separated. Therefore the total length is not always equivalent to the sum of the lengths of the head and tail.

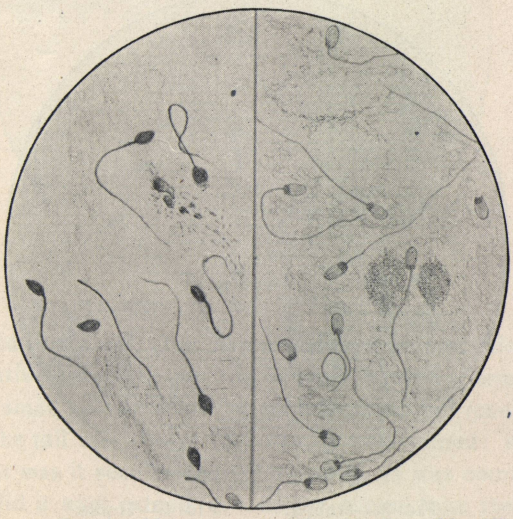


FIG. 1.—Man. FIG. 2.—Dog (Mastiff).

The spermatozoa of man (Fig. 1) were found to stain deeply with carbol fuchsin; the heads and tails were equally stained, and appeared more distinct and uniform than did those from any other member of this series. The tail was seldom found to be coiled or twisted, except where water had been previously

added to the spermatic fluid. Measurements showed the spermatozoön of man to be the smallest member of this series.

Total length,	0.051	to	0.058 mm., or 0.002	to	0.0022 in.
Length, head,	0.004	to	0.006 mm., or 0.0001	to	0.0002 in.
Width, head,	0.003	to	0.004 mm., or 0.0001	to	0.0001 in.
Length, tail,	0.041	to	0.053 mm., or 0.0016	to	0.002 in.

The spermatozoa of the dog (Fig. 2) presented many features which would readily distinguish them from those of man in the fresh and well prepared specimen; but if both were subjected to the action of certain secretions and fluids, or allowed to dry before properly smeared on the cover-glass, great question would doubtless arise as to the identity of either of these cells. A small portion of the head, located at the junction of the tail and head, stained deeply, while the remainder of the head appeared as a homogeneous structure bounded by a rather distinct margin. The tail, too, showed but moderate affinity for stains. The head and tail of an individual cell were occasionally seen to unite at right angles, and a few specimens were observed having two distinct, well formed heads projecting from a single tail. The dog furnishing the specimen for this series weighed 105 pounds, and it was the intention to compare the following measurements with those obtained by the study of spermatozoa taken from a terrier, but such opportunity did not offer itself.

Total length,	0.067	to	0.074 mm., or 0.0026	to	0.0028 in.
Length, head,	0.004	to	0.008 mm., or 0.0002	to	0.0003 in.
Width, head,	0.003	to	0.002 mm. or 0.0001	to	0.0001 in.
Length, tail,	0.059	to	0.067 mm., or 0.0023	to	0.0026 in.

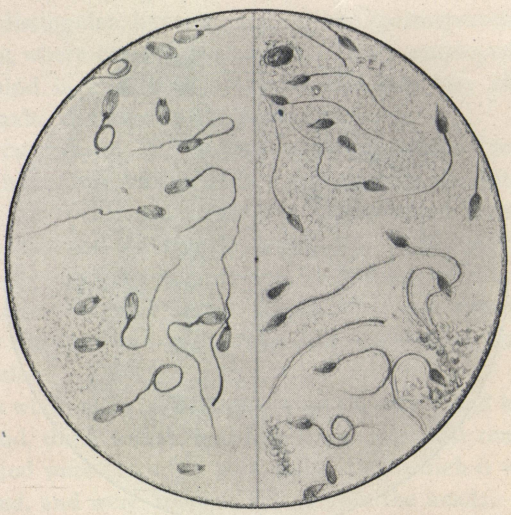


FIG. 3.—Rabbit. FIG. 4.—Horse.

The spermatozoa of the rabbit (Fig. 3) possess many features common to those of the dog; the head, in addition to being narrower, presents a deeply stained area at its junction with the tail, and a less marked deepening of the stain was seen at the other extremity, occupying nearly one-third of the whole head. Between these two stained portions a lighter zone was seen. It is

characteristic of the tail to form coils, which were often seen to surround the head, and if the smear be at all thick these coils render it impossible to outline the individual cells. An abrupt bend, at right angle, which extends for but a short distance and then forms another equally abrupt angle to assume the course previously taken, was a common finding. This irregularity in the course of the tail may be seen near the head, but more commonly at the junction of the first and second thirds.

Total length, . . 0.051 to 0.066 mm., or 0.002 to 0.0025 in.
Length, head, . . 0.006 to 0.009 mm., or 0.0002 to 0.0003 in.
Width, head, . . 0.003 to 0.004 mm., or 0.0001 to 0.0001 in.
Length, tail, . . 0.045 to 0.058 mm., or 0.0017 to 0.0022 in.

In studying the spermatozoa of the horse (Fig. 4) it was observed that their general characteristics and reaction to stain were similar to those of man, except that after the fluid had been kept for a few days the tails of certain cells appeared to be fibrillated. The measurements of equine spermatozoa were found to be:

Total length, . . 0.064 to 0.067 mm., or 0.0025 to 0.0026 in.
Length, head, . . 0.006 to 0.008 mm., or 0.0002 to 0.0003 in.
Width, head, . . 0.003 to 0.004 mm., or 0.0001 to 0.0001 in.
Length, tail, . . 0.054 to 0.060 mm., or 0.0021 to 0.0022 in.

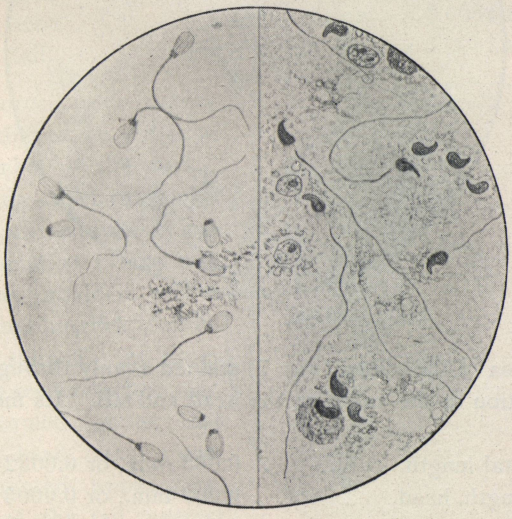


FIG. 5.—Bull. FIG. 6.—Mouse.

The spermatozoa of the bull (Fig. 5) were always accompanied by many free heads, and comparatively few free tails. The head of each spermatozoon stained feebly except for a small portion at its junction with the tail (centrosome), which stained deeply. The tail was always found to be well stained; its course rather irregular; but never was it seen to change abruptly, as was commonly observed in the rabbit, nor did it ever extend in a direct course from the head, as is the rule in man and in the horse. The measurements of bovine spermatozoa were found to be:

Total length, . . 0.087 to 0.093 mm., or 0.0033 to 0.0036 in.
Length, head, . . . 0.009 to 0.009 mm., or 0.0003 to 0.0003 in.
Width, head, . . 0.006 to 0.006 mm., or 0.0002 to 0.0002 in.
Length, tail, . . . 0.077 to 0.083 mm., or 0.003 to 0.0032 in.

The spermatozoa of the mouse (Fig. 6) presented many features in striking contrast with other members of this series. The head stained deeply and presented a slightly curved spine at one extremity, while surrounding the head was a clear zone (apparent capsule). From the portion of this capsule corresponding to the larger extremity of the head, a delicate, faintly stained tail was seen to emerge. This tail was rendered more distinct by the application of heat while staining. Measurements of these cells were found to be as follows:

Total length, . . 0.12 to 0.158 mm., or 0.0046 to 0.0061 in.
Length, head, . . . 0.008 to 0.009 mm., or 0.0003 to 0.0003 in.
Width, head, . . 0.003 to 0.004 mm., or 0.0001 to 0.0001 in.
Length, tail, . . . 0.112 to 0.138 mm., or 0.0043 to 0.0057 in.

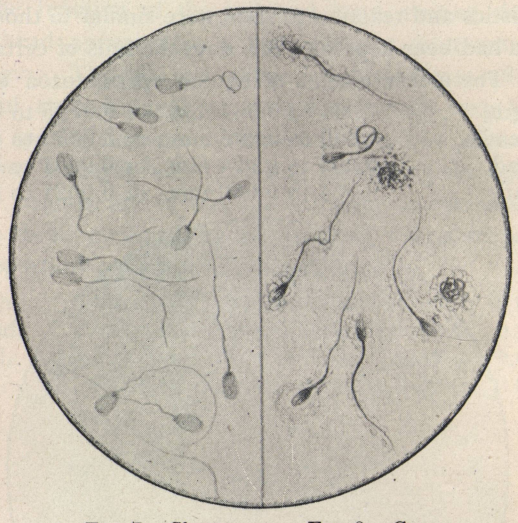


FIG. 7.—Sheep. FIG. 8.—Cat.

The spermatozoa of the sheep (Fig. 7) stained evenly throughout except for a small central portion at the union of the head and tail. Its measurements were found to be:

Total length, ... 0.083 mm., or 0.0032 in.
Length, head, . . . 0.009 mm., or 0.0003 in.
Width, head, ... 0.006 mm., or 0.0002 in.
Length, tail, ... 0.074 mm., or 0.0028 in.

Spermatic fluid from the cat (Fig. 8) was found difficult to study, as but few spermatozoa were present. Each spermatozoon presented a fibrillated tail, and at times this fibrillation was seen to surround the head. The head stained deeply, and at times a centrosome was distinct. Decided irregularity was noted in the size and form of the heads, which partially explains the variations found in the measurements of these cells.

Total length, . . 0.058 to 0.074 mm., or 0.0022 to 0.0028 in.
Length, head, . . . 0.004 to 0.007 mm., or 0.001 to 0.0002 in.
Width, head, . . 0.003 to 0.003 mm., or 0.0001 to 0.0001 in.
Length, tail, . . . 0.053 to 0.066 mm., or 0.002 to 0.0025 in.

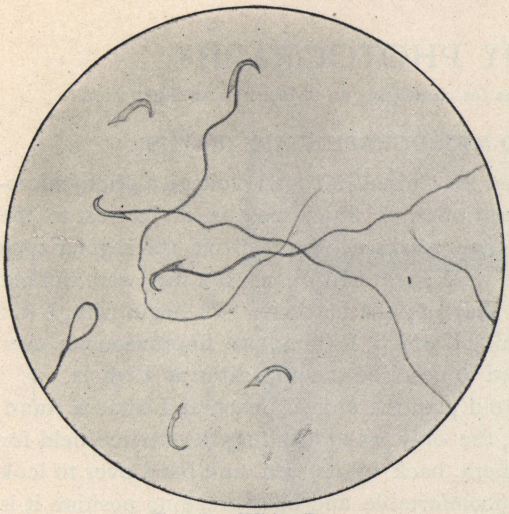


FIG. 9.—White Rats.

At first sight the spermatozoon of the rat (Fig. 9) reminds one of the immature male cell of the bird, and it is not impossible that these cells may undergo further development. The spermatic fluids of both white and gray rats were studied, and no marked difference was found to exist between these cells. The head and tail of this cell stains well, the concave border of the head staining slightly deeper than its body. Measurements were found to be:

Total length, . . 0.225 to 0.238 mm., or 0.0087 to 0.0092 in.
Length, head, . . . 0.012 to 0.016 mm., or 0.0004 to 0.0006 in.
Length, tail, . . 0.209 to 0.222 mm., or 0.0081 to 0.0086 in.

The spermatozoon of the guinea pig (Fig. 10) differs widely from any other member of the series. Its head is nearly spherical, and a minute, deeply stained portion was noted at the junction of the tail. Each head was provided with a neatly fitting, semi-lunar cap, which was also well stained. At times these caps were seen detached, and at others deformed, giving that portion of the head either a concave or a pointed appearance. That portion of the tail nearest the head was always deeply stained, and the course of the tail was never found to be tortuous. The following measurements were obtained for these cells:

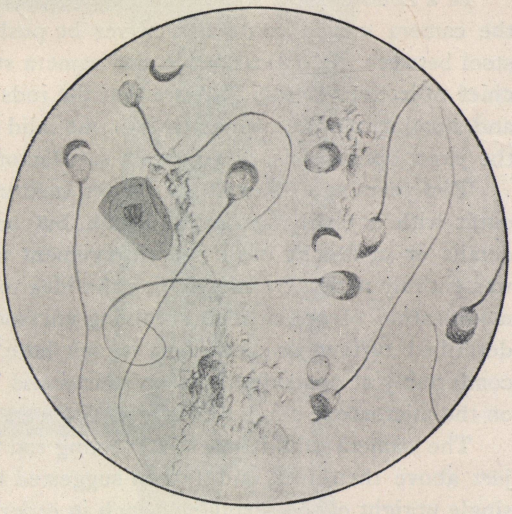


FIG. 10.—Guinea Pig.

Total length, . . 0.113 to 0.138 mm., or 0.0053 to 0.0057 in.
Length, head, . . . 0.006 to 0.012 mm.. or 0.0006 to 0.0004 in.
Width, head, . . 0.007 to 0.011 mm., or 0.0004 to 0.0004 in.
Length, tail, . . . 0.125 to 0.132 mm., or 0.0048 to 0.0051 in.

L. NAPOLEON BOSTON, M. D.

Philadelphia Hospital.

LABORATORY PHOTOGRAPHY.

Devoted to methods and apparatus for converting an object into an illustration.

AN IMPROVED PHOTO-MICROGRAPHIC APPARATUS.

I have recently had constructed for Cornell Medical College a photo-micrographic apparatus by B. & L., a description of which may be of interest to the readers of the JOURNAL, since there are certain departures from the regular type of outfit. Some of these departures were suggested by me, but they were worked out in detail and put into practical shape by the makers. To begin with, I will point out the various alterations, all of which, I think, are improvements over the old style, and afterwards proceed to describe the apparatus as a whole.

In the first place, then, on the old plan the optical bench and camera stand being practically all in one piece, the only way of finding the desired field for photographing is to push the camera back in its bed and bend over to look through the eye-piece. In this uncomfortable and back-breaking position it is impossible to manipulate the slide except by means of one of those clumsy mechanical stages which, though all very well for special purposes such as blood cell counting, are not to be compared with one's fingers, which, after all, were made at a much earlier date, for rapid manipulation.

In a Zeiss camera stand which I had previously used, the steel rods on which the camera moved could themselves be pushed back so that one could sit on a stool between the optical bench and camera stand in order to find the field. The chief objections to this plan are that the rods are apt to sag, and that the bench and stand cannot be connected into one solid piece, making it difficult to preserve the exact optical axis, or arrange a satisfactory method for mechanical focusing.

Two years ago I saw at the Jenner Institute in London a photo-micrographic outfit with a revolving optical bench, but had no opportunity of examining the details or of finding out if the arrangement worked satisfactorily or not, since it was entirely new. On mentioning the idea to Bausch & Lomb, they suggested a modification of one of their revolving microscopical tables. The details will be described further on; sufficient to say here, that by this arrangement I can sit comfortably at one side of the apparatus and with my fingers manipulate the slide on the microscope stage as easily as if I were sitting at an ordinary table.

The camera stand has a connecting rod between its two cast iron supports, just above their feet, and I then suggested that the rod be continued on to the single upright of the revolving bench in order to give more rigidity, but was told that so low down it would be of no use. After much discussion, it was decided to have a rod connecting the upper parts of the uprights, and so made that it could be put up and clamped in at the time of setting up the apparatus. This seems a very satisfactory arrangement, undoubtedly adding to the rigidity and helping to keep the optical bench and camera stand in optical axis.

The next difficulty was how to manage for mechanical focusing. When the bench and stand are fixed there is, of course, no trouble, since the focusing rod can be prolonged under the fine adjustment of the microscope, but where the bench revolves this prolongation of the rod must be got out of the way tempora-

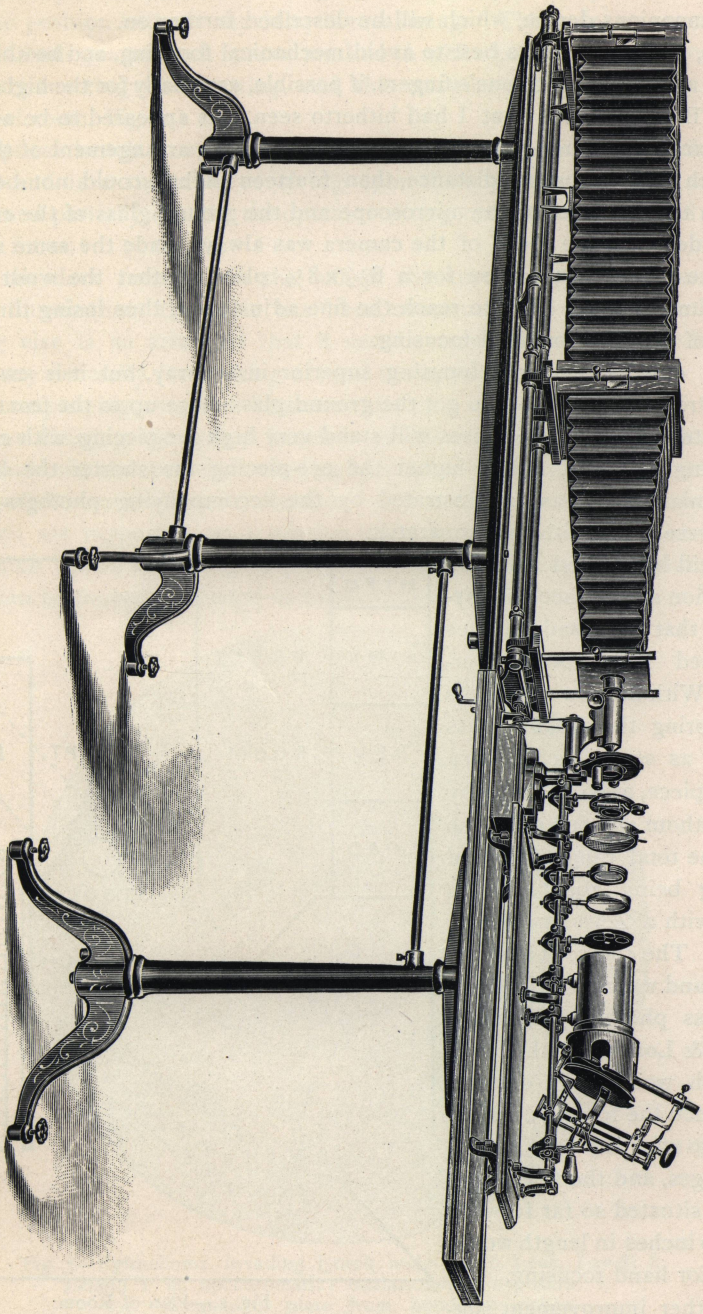


Fig. 1.—Complete Photo-micrographic Apparatus.

rily while the field is being found. Mr. Bausch asked me how I proposed to effect this, and I was forced to reply that I had not the least idea, but must leave it to him. Mr. Patterson, who had charge of the work, came to the rescue with a very ingenious device, which will be described further on.

It is, however, always best to avoid mechanical focusing, and be able to reach the fine adjustment with one's fingers, if possible, especially for the higher powers. In the B. & L. stands that I had hitherto seen, this appeared to be an impossibility, except within a very limited range, since the arrangement of the bellows was such that a shorter distance than fourteen inches could not be obtained between the eye-piece of the microscope and the ground glass of the camera.

Besides this, the front of the camera was always made the same size as the back, i. e., 11 inches square for a 6½ x 8½ plate, so that the wrist had to be bent round over the front to reach the fine adjustment, thus losing three or four inches of distance for hand focusing.

Not only is the hand focusing superior in accuray, but it is essential for another reason to be able to get the ground glass close up to the front, since the up-to-date apochromatic lenses will stand very high eye-piecing, with consequent shortening of focus. The higher the eye-piecing the shorter the focus for a given enlargement, as is illustrated by the accompanying photographs, all of which were made with this apparatus.

It will be noticed from the description and the accompanying cut that the disadvantages mentioned have been overcome. With the new apparatus I can bring the ground glass as near as seven inches from the eye-piece, and can focus by hand without any flexing of the wrist, the total range for hand focusing being about sixteen inches with a Zeiss projection ocular. The choice of a microscope stand was narrowed down to a Zeiss photographic and a Bausch & Lomb DD, the latter of which was finally decided upon since the former does not appear to offer any very special advantages, and the fine adjust ment is situated so far forward that two inches in length would be lost for hand focusing.

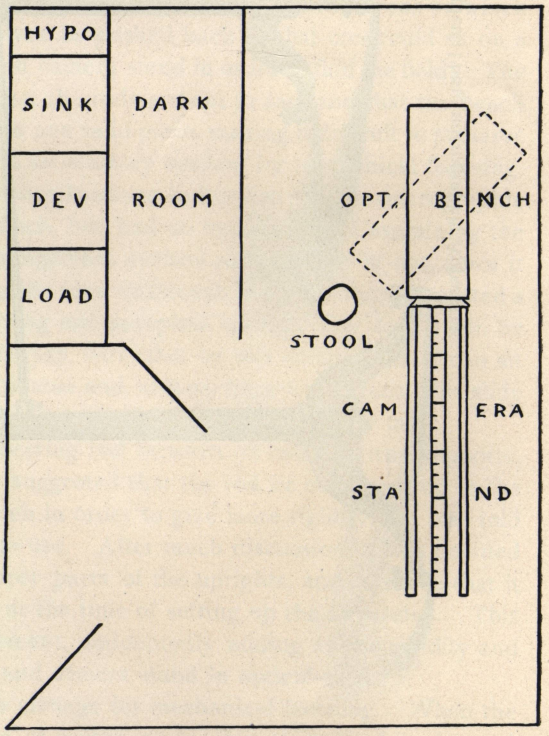


Fig. 2.—Plan of Room.

Another improvement suggested was in making the camera stand entirely of metal. The wooden bed of the old style is liable to warp, and thus throw the rods out of true.

It would have been an advantage to have had one of the rods marked off in quarter inches, so that once the length of bellows for a certain enlargement had been accurately measured, the back of the camera could at any time be brought to the same position without any delay in measuring. This point was overlooked, but I have had quarter-inch spaces marked in white paint on the flat surface of the upper connecting iron casting, and this answers the purpose just as well. (See plan of room, Fig. 2.)

The optical bench carrying lamp, microscope, and accessories is 4 feet by 15 inches and is a turntable, revolving upon the supporting column, so that it can be turned partially round; the operator sitting on a low stool to find the field and rough focus.

Since the dark room is on the left of the apparatus, looking towards the source of illumination, the table has been constructed to turn to the left, and everything else is so arranged that it can be manipulated from the left, thus obviating any necessity for walking around the apparatus.

The accompanying diagram illustrates the plan of the room, 11 x 15 feet, the dotted lines showing the optical bench in position for finding the field. Of the camera stand only the steel rods on which the camera runs, and the central beam with some of the markings, are indicated. The lamp and accessories run on steel rods, which are supported on a wooden table, supported in its turn on leveling screws, which run in a metal groove on the surface of the optical bench, so that the table can be moved backward or forward as required.

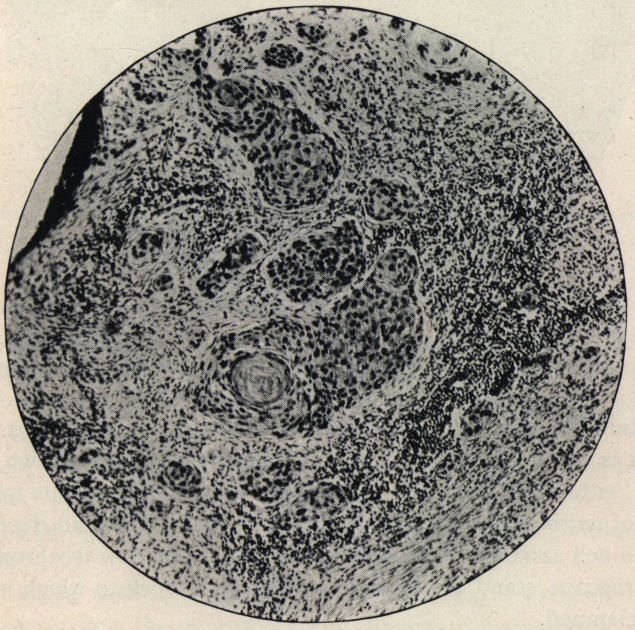


Fig. 3—Epithelioma invading lymph node. x 70. Lens, Powell & Leland 1 in. apochromatic; ocular, none; exposure, ½ sec.; distance of plate from hood, 70 inches.

The rheostat is a Colt adjustable, and stands on the floor just below the illuminating end of the optical bench. I had previously used an automatic arc

lamp, but finding that as a rule I had to be my own automaton, decided that a hand feed lamp would do better, since less liable to get out of order, and so far have no reason to regret the change.

The condenser and accessories are the regular Bausch & Lomb, so need no special description. In the figure the order from the lamp is: (1) condenser, (2) iris diaphragm, (3) paralleliser, (4) ray filter, (5) water tank, (6) iris diaphragm shutter. In practice so far, however, I have dispensed with the iris diaphragm, paralleliser, and ray filter, putting the water tank next to the condenser, and between tank and shutter using a flat tray on which ground or colored glasses, or a glass trough containing Zeltnow's solution, can be placed, so that the ray filter can be changed at a moment's notice. The lamp and condenser are then arranged so that the latter focuses directly on the substage condenser

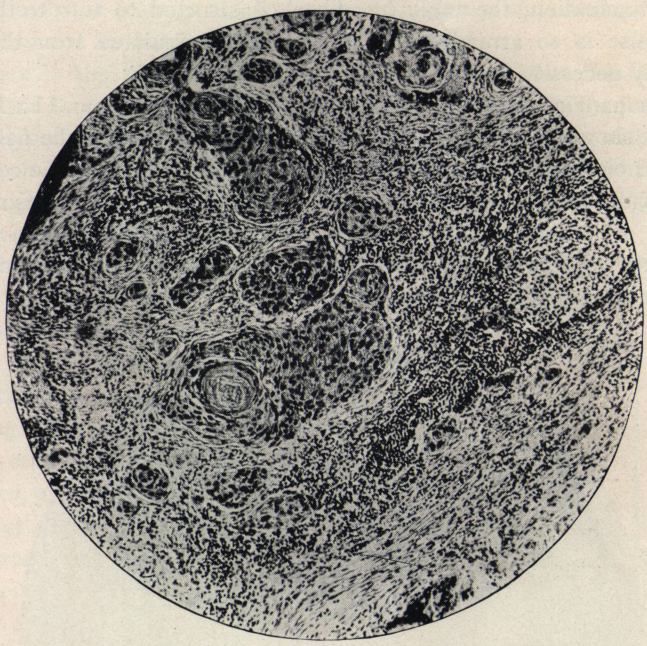


Fig. 4.—Same as Fig. 3. x70, Lens, B. & L. 1 in. apochromatic; ocular, B. & L. 1 in. compensation; exposure, 1 sec; distance of plate from hood, 12 inches.

or on the slide if no substage condenser is used. By this means a tremendous flood of light is thrown on the object, and exposures can be cut down to a fraction of a second without, so far as I can judge, affecting the results for the worse. On the day of writing this, with an exposure of one-half second, I photographed a small round cell sarcoma 250 diameters on a Cramer slow isochromatic.

The microscope stand rests on a fixed wooden block to which its horseshoe foot can be clamped.

The camera stand, as seen by the cut, consists of two cast iron uprights, connected above by a solid cast iron beam on the top of which are the already mentioned one-quarter inch spaces marked in white paint. This beam supports the steel rods upon which the camera runs.

The front of the camera is five inches square, and has vertical and horizontal movements. The bellows is divided into two, being so constructed that the back part can be taken off the central wooden frame when a short focus is required, and pushed back out of the way. The ground glass and plate-holder can then be fitted into the central frame. Both the back and central frames have vertical and horizontal movements, and are precisely alike in every particular, so that either can be used for focusing and exposing.

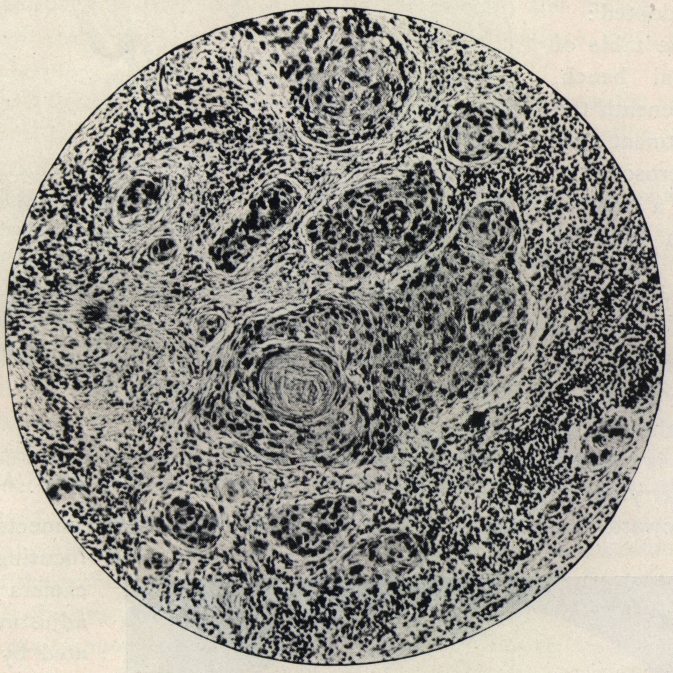


Fig. 5.—Same as Figs. 3 and 4. x 100. Lens B. & L. 1 in. apochromatic; ocular, ½ inch; exposure, 2 sec.; distance of plate from hood, 7 inches.

With regard to the arrangements for locking the optical bench, and mechanical focusing, I append a short description.

In making the mechanical attachment of a photo-micrographic camera for the purpose of operating the fine adjustment of the microscope, two points must be considered, viz. :

First. An arrangement whereby the attachment can be operated from any position, and

Second. The operation of the microscope fine adjustment without lost motion or back lash.

In the camera described above, a third problem is presented, in that the optical bench carrying the microscope and illuminating apparatus is arranged to rotate upon the supporting column, enabling the operator to adjust the specimen, illumination, and preliminary focus before connecting the microscope with the camera.

It will be seen that this arrangement necessitates the detaching of the focus-

ing rod of the camera from the microscope. To avoid the displacing and replacing of the belt connecting the mechanism, the following arrangement is adopted:

On the table of the optical bench, directly beneath the fine adjustment head of the microscope, is situated a milled wheel on suitable standard. A belt extends from this wheel to the fine adjustment head of the microscope. Through the axis of the wheel is located a rod carrying at the end toward the camera a clutch which can be quickly connected with the focusing rod of the camera by sliding adjustment, operated by a milled head. The optical bench must of course be brought to its proper position with relation to the camera in order to make this connection, and, that this position may be quickly and accurately located, an automatic catch is provided, which catch can be released by a lever, shown in the illustration.

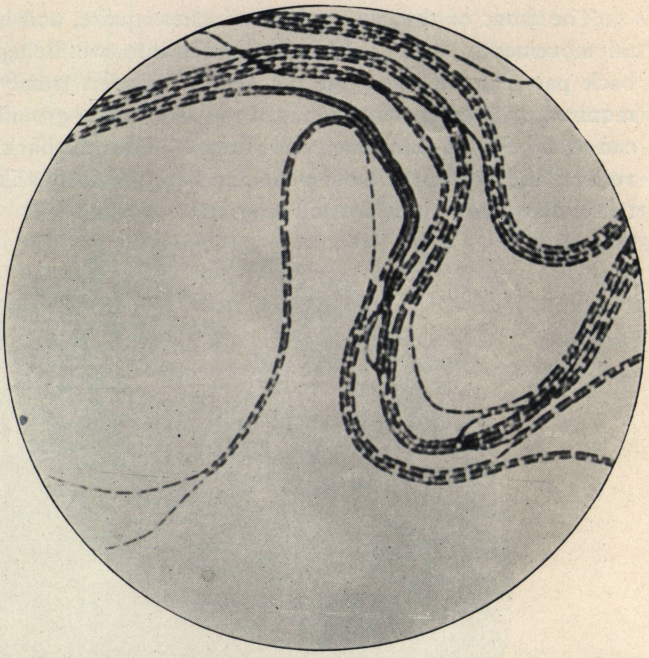
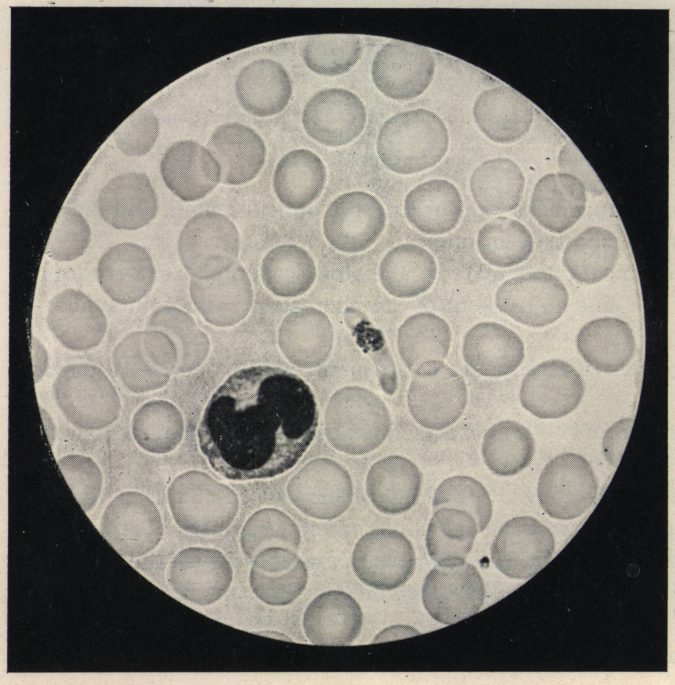


Fig. 6.—Anthrax, impression preparation from edge of colony on gelatin. x700. Lens, $\frac{1}{12}$ oil immersion; ocular 1 in. compensation; substage condenser, oil immersion N. A. 1.40.



Malarial Parasite in Human Blood. Cresentic form. x1200.

B. H. Buxton.
Cornell Medical College.

MICRO-CHEMICAL ANALYSIS.

XV.

Magnesium Group—Gl, Mg, Zn, Cd.

GLUCINUM.

This element, doubtless, should not be included in the present series of articles introductory to the methods of micro-chemical analysis, since it is rare that the analyst is called upon to search for it.

The element glucinum being, however, of much interest from the standpoint of pure chemistry, the writer has not been able to resist the temptation to include it among the few elements to be considered.

Glucinum resembles members of Group I in the crystallizing power of its chlorplatinate, this salt being analogous to that of sodium as regards its solubility and general appearance, but differs from the latter in that it crystallizes with more water of crystallization and in the tetragonal system.

Glucinum resembles aluminum and other trivalent metals in the gelatinous character of its hydroxide precipitated by ammonium hydroxide, but differs from them in that this hydroxide is soluble in solutions of ammonium carbonate.

Like magnesium, its salts unite to form double salts with ammonium; and its chloride, when evaporated to dryness from aqueous solution, is decomposed.

Like zinc, it is soluble in sodium or potassium hydroxide, the compound formed being a glucinate of the formula $Gl(OM)_2$.

It has already been seen that glucinum can replace magnesium, zinc, or cadmium in the triple acetate of sodium, magnesium, and uranyl.

It is thus obvious that in the progress of a micro-chemical analysis, glucinum, if present, may appear when testing for Group I, Group II, and, perhaps, Group III.

There are only three reagents which can be considered as giving satisfactory crystals for the micro-chemical detection of glucinum. These are:

I. Chlorplatinic Acid.
II. Normal Potassium Oxalate.
III. Uranyl Acetate and Sodium Acetate.

Of the three, the best undoubtedly is normal potassium oxalate. The other two are subject to too many disturbing conditions and sources of error.

I. Glucinum unites with Chlorplatinic Acid to form Glucinum Chlorplatinate.

$$GlSO_4 + H_2PtCl_6 = GlPtCl_6 \cdot 8H_2O + H_2SO_4.$$

Method.—Evaporate to dryness a drop or two of the solution to be tested, so as to obtain a thin, uniform film of residue. Place a drop of the reagent next to the dry residue, and carefully draw it across the latter. If the glucinum is present in considerable amount, there will appear neat, transparent, square, and rectangular plates and prisms of a faint yellow color (Fig. 64).

Remarks.—If it is desired to hasten the separation of the glucinum salt, tip up the slide and add a drop of alcohol to the test drop after the reagent has been drawn across. Generally the addition of the alcohol is essential in order that any crystals of the glucinum chlorplatinate be obtained.

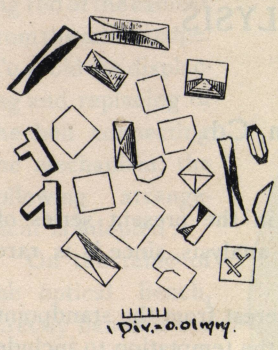


FIG. 64.

The test is satisfactory only when the air of the laboratory is quite dry. In a moist atmosphere the glucinum chlorplatinate is deliquescent, hence the test fails. In such an event it is necessary to add absolute alcohol, or place the preparation in a desiccator, or cover it with a watch-glass carrying a drop of concentrated sulphuric acid.

In case the quantity of glucinum present is small, and that of the members of Group I great, it is essential that sufficient reagent be added to unite with all. If, therefore, on examination of the preparation after the addition of the chlorplatinic acid, it is seen that members of the potassium group are present, it is wise to make a second addition of the reagent, and follow it with alcohol.

When sodium is present in considerable amount, it is often difficult to distinguish the glucinum salt from the sodium chlorplatinate; if, however, the preparation be examined between crossed nicols, the problem is simplified, since the chlorplatinate of sodium exhibits oblique extinction (triclinic) and a brilliant play of colors, while the glucinum compound gives parallel extinction (tetragonal) and but faint colors (usually none). The chlorplatinates of the potassium group are isometric.

If solutions containing glucinum in the form of sulphate are employed, care must be taken to avoid confusing this salt with the chlorplatinate, since the glucinum sulphate, $GlSO_4 \cdot 4H_2O$, which is also to be referred to the tetragonal system, sometimes separates in thin, six-sided plates.

II. Normal Potassium Oxalate added to solutions of salts of Glucinum causes the separation of a difficultly soluble Double Oxalate of Potassium and Glucinum.

$$2\,K_2C_2O_4 + GlSO_4 = K_2C_2O_4 \cdot GlC_2O_4 + K_2SO_4.$$

Method.—To the moderately concentrated solution add a little acetic acid, then a fragment of the reagent about twice as large as is usually the case in micro-chemical work. Almost immediately large, clear, colorless, highly refractive prisms of the monoclinic systems are obtained. These prisms unite to form twins and radiating masses (Fig. 65).

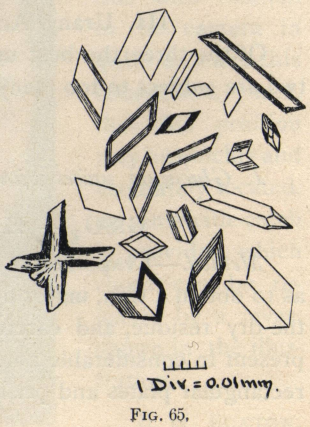


FIG. 65.

Remarks.—The appearance of the crystals varies greatly, according to the amount of the reagent present, as compared with that of glucinum. Too little potassium oxalate will yield only a precipitate of tiny crystals which probably consist of the normal oxalate of glucinum. Too much re-

agent, on the other hand, gives rise to skeleton crystals and to masses of thin lenticular plates.

During the disintegration of the fragment of potassium oxalate while passing into solution (particularly in concentrated solutions), crystals of the reagent appear *momentarily*, which bear a striking resemblance to some of the forms assumed by the double glucinum potassium oxalate. In testing unknown solutions the worker must be on his guard lest he fall into error by deciding too hastily.

The double oxalate of glucinum and potassium can be readily recrystallized from water by gently warming the preparation and allowing it to cool slowly. The salt is also soluble in solutions of ammonium carbonate, a property which can be utilized when there is doubt as to the nature of the precipitate obtained in the course of an analysis.

The addition of a little mercuric chloride will induce the production of long prisms and twins, and hence is useful when good crystals cannot otherwise be obtained.

Neither primary potassium oxalate, sodium, nor ammonium oxalates can be substituted for the normal oxalate of potassium.

With zinc, the reagent gives tiny double globulites and pseudo-octahedra of normal zinc oxalate, and later, as the test drop concentrates by evaporation, neat hexagonal plates appear, which are probably due to a double oxalate of potassium and zinc (?). Mercuric chloride seems to favor the formation of the hexagonal plates.

Cadmium treated in like manner yields, apparently, only crystals of normal cadmium oxalate (g. v.). No double salt seems to separate.

The reagent gives nothing with magnesium, providing the test drop is not too concentrated and does not contain an excessive amount of free acetic acid.

When zinc or cadmium is also present, the crystal form of the glucinum potassium oxalate is changed. It then becomes difficult to decide whether or not glucinum is present.

Magnesium, aluminum, and iron, on the other hand, have practically no influence, unless present in relatively large amount. But the double oxalate of glucinum and potassium crystallizing from such solutions will always occlude an appreciable quantity of the potassium double oxalate of these elements.

Calcium, strontium, and barium may mask the reaction.

Ammonium salts, if present, must first be removed by gentle ignition before testing with potassium oxalate.

Free mineral acids must be absent.

Stannous salts may at times give, with potassium oxalate, crystals of stannous oxalate which may be mistaken by an inexperienced worker for the glucinum double salt.* After the tin salt has been allowed to grow for a short time, there is little danger of confusing the two. If still in doubt, recrystallize from warm water, treat with ammonium carbonate, or apply tests for tin.

When the solution to be tested contains copper, cobalt, or nickel, it is gener-

* This error cannot arise in the course of a systematic analysis.

ally best to avoid testing it directly for glucinum, but to first practice a separation where the former are removed from the latter.

Exercises for Practice.

To a drop of a solution of a pure salt of Gl add $K_2C_2O_4$ in the manner directed above. Try the experiment several times, varying the amount of the reagent.

Try again under as nearly like conditions as possible, but this time having first introduced a little $HgCl_2$.

Try the action of HKC_2O_4; $(NH_4)_2C_2O_4$; $Na_2C_2O_4$.

Try reagent on salts of Mg; Zn; Cd; Cu; Co; Ni.

Then try mixtures, as for example, Gl and NH_4; Gl and Mg; Gl and Al; Gl and Fe; Gl and Zn; Gl and Ca, etc., and also more complicated mixtures.

III. With Uranyl Acetate and Sodium Acetate.

This reaction of glucinum salts has already been alluded to under Sodium, Method II.* The equation for the reaction is the same as that there indicated, save that glucinum replaces the magnesium.

To the material to be tested a little sodium acetate is added (unless it is known that sodium is present). The drop is then evaporated to dryness, and the solution of the reagent is drawn across the film of dry residue. Skeleton crystals, long, imperfect prisms, and almost colorless tetrahedra result.

There is, at times, some difficulty in clearly distinguishing between the triple acetate of glucinum, sodium, and uranyl, the double acetate of sodium and uranyl, and the crystals due to a separation of uranyl acetate. It has been the experience of the writer that students trying the method for the first time are invariably in doubt as to the nature of the crystals obtained.

The tetrahedra of the triple acetate differ only in size and color from those of the double acetate, the former attaining a greater size than the latter, and being only very faintly yellow instead of exhibiting a distinct yellow tint.

The amount of sodium present must be small, otherwise only the double acetate will appear.

Salts of ammonium, potassium, and of the calcium group will generally interfere, if present in excessive quantity.

Phosphates and other compounds precipitating uranium should be absent.

Free mineral acids must be removed by evaporation to dryness, as has been suggested.

It is obvious that this method cannot be employed for the detection of glucinum save in the absence of magnesium, zinc, cadmium, cobalt, nickel, iron, manganese.

MAGNESIUM.

The micro-chemical detection of magnesium in complex mixtures is usually a matter of not a little difficulty, since this element is commonly associated with others closely related, which are prone to interfere with or prevent the formation of typical crystals with the reagents employed for its recognition.

* Jour. App. Micros. III, 1900, 985.

Tests applied to pure salts and simple mixtures are quite satisfactory, and would scarcely lead the worker to anticipate the annoyances and difficulties which may beset him in other cases.

In ordinary practice three reagents will be found useful:

I. Secondary Sodium Phosphate in Ammoniacal solution.
II. Potassium Antimonate,
III. Uranyl Acetate with Sodium Acetate.

I. The addition of Secondary Sodium Phosphate to Ammoniacal solutions containing Magnesium precipitates Ammonium Magnesium Phosphate.

$$MgSO_4 + HNa_2PO_4 + NH_4OH = NH_4MgPO_4 \cdot 6H_2O + Na_2SO_4 + H_2O.$$

Method.—Two methods are available; the choice of procedure depending upon the nature of the salts present in the drop to be tested. In all cases where there is a doubt as to the probable composition of the material to be examined, it is best to have recourse at once to the modification *B*.*

A. To the solution of the material to be tested, which must not be too concentrated, add several fragments of ammonium chloride; stir, then a very slight excess of ammonium hydroxide, and warm the preparation. (If a precipitate results it is best to draw off the clear solution.) To the warm solution add a small crystal of secondary sodium phosphate. Crystals of ammonium magnesium phosphate soon appear.

B. To the solution to be tested add a fragment or two of citric acid, then an excess of ammonium hydroxide. Evaporate to dryness. To the residue add dilute ammonium hydroxide. Warm, then add a very little solid secondary sodium phosphate. Crystals of ammonium magnesium phosphate separate.

The crystals of the ammonium magnesium phosphate separate as skeletons and hemimorphic forms of the orthorhombic system (see Figs. 40 and 66).

Remarks.—It should be remembered that a number of elements are precipitated by phosphates in alkaline solution; the most frequently met with in the course of micro-chemical analyses, either in the substance to be tested, or present as reagents from previous tests, are doubtless, lithium, members of the calcium and magnesium groups, trivalent metals, manganese, nickel, cobalt, tin, lead, silver, copper, uranium.† Of these elements, lithium, iron, manganese, cobalt, and nickel form, with ammonium and phosphoric acid, salts of similar composition to and isomorphous with the magnesium salt.

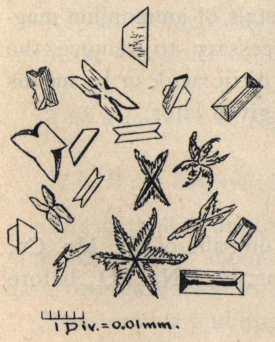


FIG. 66.

The ammonium glucinum phosphate, ammonium zinc phosphate, and ammonium cadmium phosphate are not precipitated in crystal form.

* Romijn, Zeit. anal. Chem. 37, 300.

† Most of these elements will have been removed in the progress of the analysis before the addition of the sodium phosphate.

In *A* the reaction sometimes fails for lack of sufficient ammonium chloride, magnesium hydroxide being precipitated. A slight excess of this salt will do no harm.

Both modifications fail if there is an insufficiency of ammonium hydroxide, for it should be remembered that there must be not only enough ammonium present to unite to form the proper compound, but that this salt will not separate save in alkaline solution.

The advantage of employing modification *B* lies in the fact that owing to the presence of ammonium citrate, there is little danger of the interference of the elements listed above. If, in following this method, the residue after evaporation is not completely soluble in the ammonium hydroxide solution, it is best, though not essential, to draw off the clear liquid before adding to it the sodium phosphate.

Reactions *A* and *B* work equally well in the cold, but are then a trifle slower. Generally, an amorphous precipitate is at first produced, which begins to crystallize in a few seconds. The formation of merely an amorphous precipitate must never be taken as evidence of the presence of magnesium.

It must also be borne in mind that the use of too strong ammonium hydroxide in excess so reduces the solubility of many salts as to cause their separation, hence it is necessary to beware, in reactions of this character, of deciding too hastily as to the result of a test.

See remarks made under Ammonium, Method II (JOURNAL, p. 1190), and Calcium, Method V (JOURNAL, p. 1247).

In the presence of phosphates the detection of magnesium becomes quite difficult, particularly if other elements are present which form phosphates insoluble in ammonium hydroxide. If arsenates are also present, a still further com plication arises, for, as we have already seen, double ammonium arsenates of calcium, zinc, etc., are formed, which are isomorphous with ammonium magnesium phosphate.

Of course it may happen that in some cases the mere addition of ammonium hydroxide will cause the separation of characteristic crystals of ammonium magnesium phosphate. Generally, however, it is first necessary to remove the phosphoric acid. This can be accomplished by tin and nitric acid, or by means of ammonium tungstate and nitric acid. Details will be given later.

Exercises for Practice.

Try method *I A* on a solution of $MgSO_4$, then try it on salts of Fe, Mn, Co, Ni, Al, Zn, Cd. Repeat the experiments, this time adding the HNa_2PO_4 before the NH_4OH.

Try *I B* in like manner.

Make mixtures, trying various combinations of the above with members of Groups I and II.

Consult notebook on the results obtained with the experiments tried under Ammonium II and Calcium V.

II. Potassium Antimonate added to solutions containing Magnesium causes the separation of Magnesium Pyro-antimonate.

$$MgSO_4 + H_2K_2Sb_2O_7 = H_2MgSb_2O_7 \cdot 9H_2O + K_2SO_4.*$$

Method.—First prepare an almost saturated solution of the reagent by heating a fragment with water. A drop of this solution is placed next the test drop, and the two caused to unite. A dense amorphous precipitate is usually immediately produced. After a time, crystals of magnesium pyro-antimonate appear, generally near the circumference. The forms most frequently obtained are thin, colorless, transparent hexagonal plates, and spherical masses more or less crystalline in appearance (Fig. 67). Less often, short hexagonal prisms are seen.

Remarks.—The solution to be tested must be dilute and neutral. Free acid not only interferes with the formation of characteristic crystals, but also causes the reagent itself to yield an amorphous precipitate.

The development of the crystals of magnesium pyro-antimonate is quite slow, and eventually they may attain a size of double or even triple that of those shown in Fig. 67.

Alcohol can be employed to hasten crystallization, but it is better to allow the preparation to take all the time it needs.

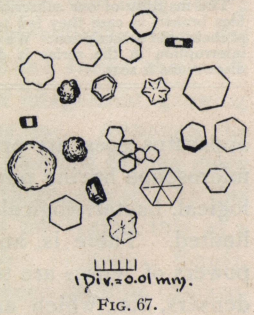


FIG. 67.

Lithium sometimes yields crystals not to be distinguished from those of magnesium, more often circular disks and sperulites.

Sodium (q. v.) gives fusiform crystals.

Members of the calcium group are precipitated in an amorphous form, and interfere with the test for magnesium.

Ammonium salts should be absent.

Exercises for Practice.

Try reaction on salts of Mg.

Repeat the experiment in the presence of salts of NH_4.

Make a mixture containing Na and Mg; test as above.

Test a salt of Li. Try the effect of the reagent on salts of Zn and of Cd.

Test a mixture of Mg and Zn.

III. With Uranyl Acetate and Sodium Acetate.

The method of applying the test has been described in Method III of Glucinum; there, and under Sodium, Method II, the properties of the triple acetates have been discussed in detail.

The formula and appearance of the triple acetate of sodium magnesium and uranyl will be found on page 985 of Vol. III, of this Journal. To this article, and to that on Glucinum, the reader is referred for details as to methods of procedure, sources of error, etc.

E. M. CHAMOT.
Cornell University.

* See foot note, Sodium, Method V, Jour. App. Micros. III, 1900. p. 1048.

Journal of
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and
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Edited by L. B. ELLIOTT.

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AMONG the many questions clamoring for decision, that of the standard of equipment in the various classes of laboratories has received very little organized consideration, and yet it is one of as great practical value as any connected with science teaching, and one which would seem to admit of a very easy and practical settlement. It is not our purpose here to make any suggestions as to what might constitute a standard equipment, but to point out the value to educational institutions of adopting a standard.

The requirements for each class of laboratory, so far as the most important apparatus goes, are practically the same. The biological laboratory requires a microscope having powers ranging from a minimum to a maximum. The histological, bacteriological, chemical, high school, and each other class is likewise limited. There is, however, no unity of opinion as to the kind of stand these powers of lenses are to be used on, or the accessories, such as nosepiece, condensers, etc., which are to be used with them. The same is true in a general way in regard to microtomes, incubators, and the unit equipment, for each student, of glassware, stains, and reagents. Each laboratory director is a law unto himself, and an inspection of the purchases made for the various laboratories of the country for the year would seem to indicate that each had done his best to be original in the make-up of his equipment.

This is all well enough from the standpoint of the individual who is equipping the laboratory, but the practice costs the institutions of the country an immense sum of money, far greater than any one not fully conversant with the conditions can realize, and the cause is obvious.

The cost of any article is dependent very largely on the number consumed. Where the number is small the cost of production is high, because it does not pay the manufacturer to build expensive machinery and make up a large quantity to be held a long period, and in addition run the chance of his stock becoming antiquated through the development of more suitable models.

So with this laboratory apparatus; the ceaseless demand for variations from existing models, the selection of every grade of apparatus for the same kind of work, makes it necessary for the maker to build an endless variety of perfectly useless instruments, and to charge an average advance on all to compensate him for the extra cost to him of doing his work piecemeal.

If the subject of laboratory equipment could be taken up by a committee from each of the organizations of laboratory men interested in this work, and a joint recommendation made, there is no question but that the majority of laboratories would accept the findings, and that the uniform demand thus established would result not only in better apparatus, but at a decreased cost.

CURRENT BOTANICAL LITERATURE.

CHARLES J. CHAMBERLAIN.

Books for review and separates of papers on botanical subjects should be sent to Charles J. Chamberlain, University of Chicago, Chicago, Ill.

REVIEWS.

Murbeck, S. Ueber das Verhalten des Pollenschlauches bei Alchemilla arvensis und das Wesen der Chalazogamie. Lunds Universitets Arsskrift. **36**: 1–19, pls. 1–2, 1901.

Since other species of Alchemilla have been found by the same writer to be parthenogenetic, it is of interest to note that *A. arvensis* has a pollen tube at all. In the development of the ovule a micropyle is formed, but long before pollination occurs the continued growth of the integument entirely closes the micropyle. The pollen tube grows down through the style and enters the ovule at the chalazal end, then traverses the entire length of the integument, growing through the tissue, and enters the micropylar region of the sac. Although the act of fertilization was not observed, it may be reasonably assumed to take place in spite of the fact that other species of *Alchemilla* are parthenogenetic. In this case the pollen seems perfectly normal. The author does not regard this as a case of chalazogamy such as is found in *Casuarina*, *Corylus*, *Carpinus*, *Betula*, *Alnus*, and *Juglans*, but rather as a type intermediate between genuine chalazogamy and the condition found in *Ulmus*. Chalazogamy is regarded as a derived condition, and as a physiological phenomenon of no phylogenetic significance.

C. J. C.

Brand, F. Bemerkungen über Grenzzellen und über spontanrothe Inhaltskörper der Cyanophyceæ. Ber. d. deutsch. bot. Gesell. **19**: 152–159, 1901.

The heterocysts of the Nastocaceæ have been described as cells poor in contents, or with only watery contents, and they have been supposed to be concerned only in false branching and in breaking filaments up into hormogonia. The present writer, in investigating *Nostoc commune*, finds that in addition to the empty heterocysts there are also heterocysts with contents, which are not watery but elastic, and of considerable consistency. By pressure on the cover-glass, the walls of these heterocysts may be broken and dissociated from the contents which retain their spherical form. It was found that the contents divide like ordinary vegetative cells, and give rise to filaments. It was also found that in some cases the contents of the heterocyst pass over into the neighboring cells, and may induce in them a renewed activity. The writer believes that the red granules of the Wasserblüthe, forming members of the Cyanophyceæ, are not due to gases.

C. J. C.

Ernst, A. Beiträge zur Kentniss der Entwickelung des Embryo-sackes und des Embryo (Polyembryonie) von Tulipa Gesneriana L. Flora. **88**: 37–77, pls. 4–8, 1901.

This paper treats in considerable detail the life history of *Tulipa Gesneriana*, from the appearance of the archesporial cell in the nucellus of the ovule up to the ripe seed. Besides the original work, there is a very convenient summary of the literature of polyembryony. A few of the points noted are the following:

The first division of the nucleus of the embryo-sac in which the reduction in the number of chromosomes is effected, takes place after the opening of the flower. The number of chromosomes in the gametophyte is twelve, but in one case six were counted. The antipodal nuclei become fragmented into a varying number of pieces. The generative cell of the pollen grain often occupies the greater portion of the space within the spore, and has an unusually thick membrane. The vegetative nucleus remains in the end of the tube after the two male nuclei have been discharged into the sac. One of the male nuclei conjugates with the nucleus of the egg, and the other becomes applied to the upper polar nucleus, so that the definitive nucleus results from the fusion of three nuclei, the two polar nuclei and one of the male nuclei.

After fertilization, the egg gives rise to an irregular mass of cells, in which the beginnings of several embryos may be distinguished. The polyembryony of *Tulipa* is very much like that described by Jeffrey for *Erythronium*.

C. J. C.

CYTOLOGY, EMBRYOLOGY, AND MICROSCOPICAL METHODS.

AGNES M. CLAYPOLE, Cornell University.

Separates of papers and books on animal biology should be sent for review to
Agnes M. Claypole, 125 N. Marengo avenue,
Pasadena, Cal.

CURRENT LITERATURE.

Doflein, F. Cell Division in Protozoa. Zool. Jahrb. **14**: 1–16, 1900. Extracts from Royal Mic. Jour. April, 1900.

Dr. F. Doflein has studied *Noctiluca miliaris* with especial reference to the nuclear changes accompanying cell division. The life cycle is as follows: After dividing repeatedly the adult comes to rest, copulation of two individuals occurs, followed by rapid budding. The liberated buds are at first similar to Dinoflagellata, but ultimately become converted into adults. Division occurs, a sphere appears near the nucleus, and a process takes place believed by the author to have a superficial resemblance to metazoan karyokinesis. The division of the nucleus appears to be in some degree independent of the division of the sphere, the division of the latter being closely associated with plasmic division; the author believes that the former structure is but a concentration of the plasma itself, hence the close relation. The budding after copulation consists of a rapid cell division during which the division products remain united by a common stroma. It is uncertain whether this stroma indicates a reduction process or not. A discussion on the structure of protoplasm and the causation of its movement is also given in the paper.

A. M. C.

Stephens, J. W. W. and Christopher, R. S. R. Technique for Malaria Blood. Roy. Soc. Report to Malaria Comm. 3d Series, 1900. Ext. from Royal Mic. Jour., April, 1900.

The authors use the following simple method for preparing and staining films of malaria blood: The finger is pricked with a triangular surgical needle and a clean glass slide touched to the exuding blood. The drop thus obtained

on the slide is spread by the shaft of the needle in a broad, even streak, a little time being allowed for the drop to run along the needle by capillarity. The most perfect films are thus obtained. The slides are then placed in absolute alcohol for 5 minutes, after which the films are stained with saturated alcoholic solution of hæmatein. To every 10 c. c. of this solution is added 50 c. c. of alum solution (alum 50 grams, water 1000 c. c.). In this solution the slides are left 5 to 20 minutes or even hours. Oil is applied directly to the slide without a cover and the specimen examined. A permanent mount can be made by washing off the oil with xylol and mounting the preparation in balsam. If placed in a clean box and wrapped in paper, the slides will keep a year unmounted. A. M. C.

Gurwitsch, A. Die Vorstufen der Flimmerzellen und ihre Beziehungen zu Schleimzellen. Anat. Anz. **19**: 44–48, 1901.

In the eighteenth number of this periodical a work on M. Heidenhain appeared which gives an opportunity for a few remarks. Heidenhain's work came to the author so shortly before the appearance of his previous communication on this subject that it was impossible to discuss in the same issue Heidenhain's opinions of the author's statements. These are here set forth. The peculiarly shaped epithelial cells in the mouth and pharynx of salamander larvæ is the subject under special discussion.

The author has presented, beginning with the earliest stages, the development and its modifications of the peculiar superficial border of the cells; first appearing as an apparently homogeneous "crest" not sharply separated, the cell border is next clearly foam-like; in the course of the further development the foam-like structure is effaced to make a "felt work." (Arch. f. Mikros. Anat. **57**: 209, Fig. 16–18.)

As an end product of development there comes a clearly formed, sharply isolated border of small rods, of which the separate little hairs correspond in their height exactly to the cilia of the mature ciliated cells and provisionally remain covered with a thin but very sharply apparent, net-like film. At this stage of development direct observation ceased, as the oldest of the remaining larvæ showed no more continuance of the process. Based on this observation the author felt drawn to the conclusion that these cells possessed of rods were the early stages of ciliated cells, and also to infer that in one kind of cells at least the cilia are formed before the basal bodies.

This interpretation of these questioned cells is now doubted by M. Heidenhain. He regrets that the "last step of development, the peculiar transformation into the true, mature, free cilia, was not observed." The reason for this deficiency was given thus: "I sought to remedy the lack somewhat by figuring in my detailed work (Fig. 21) a mature ciliated epithelial cell next to two intermediate ones from the transition of esophagus to pharynx." Heidenhain is much more disposed to consider the pharyngeal cells as forerunners of mucous cells. It seems that a misunderstanding arises each time. Although in both articles the authors speak only of pharyngeal, not of esophageal cells, and although Heidenhain mentions each time that the pharyngeal epithelium of the salamander was the object treated of, he suggests as a possible cause for confusion on

Gurwitsch's part the circumstance that "the epithelium of the esophagus and stomach in the transition region may not be sharply distinct from each other, so that ciliated regions could be found in the surface epithelium." This is impossible; since the part of the epithelium which was used for investigation lay above the esophagus, and since the whole region was covered with a similar unbroken coat of these questioned cells, just as in later life the ciliated coat is entirely unbroken, the author's conclusion is again justified and Heidenhain's assumption would only be right if there were ground for the belief that the whole pharyngeal epithelium was changed to a mucous condition and that later ciliated epithelium arose *de novo* from some unknown source. No evidence exists for this and such a process could not easily escape notice. Moreover, if Heidenhain's explanation were applied to all these questioned cells, then this rodded border, which occurs in so many forms and kinds of cells, must be declared the forerunner of *mucoid* formation on the ground alone that it seems to be the case in this one kind of cell.

It cannot be doubted that identical tissues in two nearly related species of animals have in similar developmental stages an entirely different appearance. Therefore it is no objection to the writer's hypothesis that the methods of development of ciliated cells in salamanders may differ in various cases. The only apparent question existing is whether these steps are those of ciliary or mucoid formation in the cells.

The facts important for histogenesis in general must satisfy us that similar structures may owe their origin to different methods of development, and that the histogenetic processes which are found in one species may not be applied to a nearly related one. This is also true in other lines, as for example, the histogenesis of crystalline lens in different animals and the ectodermal origin of cartilage in Petromyzon.

The author adds a few words on the beaker cells in the same epithelium. This in the salamandar larva has always two layers, the one with the peculiar rod-cells is set on a layer of cubical or more or less sloping cells. In not a single case was the first or rodded layer found in contact with the basilar membranes. On the other hand, all the mucous cells, independent of their condition of function, were placed directly on the basilar membrane. This immediately suggested the latter to be the mother cells of the beaker cells. While this could not be maintained by direct evidence, not sufficient transitional stages being examined, it is enough for this argument to state the facts that there is a good criterion for the differentiation of the immature mucous cell from the rod cell. "Thus the beaker or goblet mucous cells of the pharyngeal epithelium point clearly to a layer underlying the latter ciliated cells.) This relation holds true in all cases and all stages in development. E. J. C.

CURRENT ZOÖLOGICAL LITERATURE.

CHARLES A. KOFOID.

Books and separates of papers on zoölogical subjects should be sent for review to Charles A. Kofoid, University of California, Berkeley, California.

Bergh, R. S. Kleinere histologische Mittheilungen, Zeitsch. f. Wiss. Zool. **69**: 444–456. Taf. 32, 33, 1901.

The author commends the use of maceration methods in the study of the histology and organology of the larva of the leech *Aulastoma*. Very dilute acetic acid or a mixture of three or four parts of 30 per cent. alcohol with one part of 2 per cent. acetic acid was employed with good results, both for maceration and as a fluid for examination. For the demonstration of cell boundaries the silver method of Fischel was used, though the finest results were secured with a mixture of equal parts of 1 per cent. nitric acid and 1 per cent. silver nitrate allowed to act for a longer time than that usually employed in silver impregnation. Reduction was accomplished by sunlight or by a weak solution of formic acid in alcohol. By this method very fine demonstrations of the cell boundaries in the nephridia of the *Lumbricidæ* can be secured, the cell limits being defined even in the intracellular lumen. Bergh confirms his earlier thesis of the presence of a larval epidermis in *Aulastoma* consisting of about thirty large multinucleate cells, whose nuclei multiply by amitotic division. The structure of the nephridium of *Lumbricus herculeus* is considerably elucidated by the silver method. The cells in the margin of the funnel alone have the usual straight cell walls. Within the funnel and in the straight, ciliated, intracellular lumen of the adjacent section of the nephridium the cell walls are very tortuous. In the narrower regions when the lumen is intracellular they become even more irregular and in the ampulla almost labyrinthine, though everywhere transverse in general direction, each cell forming a short transverse section of the nephridial tube. Attempts to demonstrate cell boundaries in the nephridia of the aquatic oligochætes by the silver method failed entirely. C. A. K.

Coe, W. R. Papers from the Harriman Alaska Expedition, XX. The Nemerteans, Proc. Wash. Acad. Sci. **3**: 1–110, pl. 1–13, 1901.

Dr. Coe reports thirty-two species, of which all but two are new to the Pacific region and twenty-seven new to science. The methods employed with this refractory group are of general interest. The worms die well extended if a few drops of formalin are added to the sea water in which they are placed, and if handled with care they do not always break up into fragments. Material was hardened in 2 to 5 per cent. solution of formalin in sea water and eventually transferred to alcohol. Formalin gives good results for anatomical work or for the histology of epithelial structure, but it is disastrous to the connective and nervous tissues. For supplementary work, strong alcohol, sublimate-acetic, Gilson's fluid, and—for nervous system—Flemming's fluid were used. Iron hæmatoxylin followed by orange G was the most effective stain for sections. C. A. K.

Burckhard, G. Die Implantation des Ei der Mans in die Uterusschleimhaut und die Umbildung derselben zur Decidua. Arch. f. Mik. Anat. u. Entwick, **57**: 528–569, Taf. 26–28, 1901.

It was the purpose of this investigation to follow the changes in the uterus from the time the egg enters it from the oviduct until the embryo is fully encapsuled in its walls in the so-called *decidua reflexa*. About the beginning of the fifth day after impregnation in the mouse, the ova are clustered at the lower end of the oviduct in an advanced stage of cleavage with a small cleavage cavity appearing. About this time they enter the uterus and are immediately distributed, probably by movements of the uterine walls, at somewhat regular intervals throughout the uterine lumen, lying in crypt-like depressions on the antimesometrial side of the lumen. The process of implantation is completed by the eighth day and, owing to the rapidity with which it takes place, has been overlooked in large part by all previous investigators of the subject. At the end of the eighth day the embryo is separated from the lumen and embedded in the decidua, composed entirely of mucosa cells from which all traces of the *uterine epithelium have disappeared*. Other investigators have suggested that the embryo sinks beneath the epithelium and develops in the mucosa, but Burckhard's results show conclusively that this position of the embryo is brought about by a *degeneration* of the uterine epithelium of the walls adjacent to the embryo. By the middle of the fifth day the epithelium near, but not as yet in contact with, the ovum, shows traces of flattening and the cells of the subjacent mucosa exhibit nuclear activity. Eosinophilous leucocytes invade this territory and the capillaries branch and spread toward the region of the ovum, while the uterine glands close and degenerate from the uterine lumen toward the musculature. By the middle of the sixth day the epithelium near the ovum (lining of the decidual cavity) is much flattened and the walls on the mesometrial side meet above the ovum uniting with its ectoplacental one (Träger), thus completely separating the decidual cavity from the uterine tract. By this time the epithelium near the ovum has disappeared entirely either by degeneration or retraction, while the remainder of the lining of the decidual cavity degenerates by evident desquamation and karyolysis.

The ovum now lies in the uterine mucosa beneath the epithelium on the antimesometrial side of the uterus. Multiplication of the decidual cells and their subsequent growth, combined with increased vascular supply, result in further closure of the uterine walls above the ovum until only a small lumen remains. The thickened vascular region above the ovum becomes the placenta, while the persistent lumen, now on the mesometrial side of the ovum, comes at a later stage to lie on the antimesometrial side. The manner in which the change is accomplished is not at present known. The probability of a method of implantation of the ovum in the human uterus similar to that found in the mouse is suggested and discussed at length. The material examined was secured entirely from white mice. The uteri were removed immediately after death to picro-sublimate, Zenker's, Flemming's or Hermann's fluids for twenty-four hours. Safranin iron-hæmatoxylin was used after the last two fluids, borax carmin or hæmatoxylin-eosin after the other fluids. These last preparations gave the best results; since the eosin differentiates the blood corpuscles and also gives the epithelium of the uterus and its glands a characteristic tint. The figures illustrating this paper are very fine, being based upon micro-photographs prepared by Sobotta's method. C. A. K.

NORMAL AND PATHOLOGICAL HISTOLOGY.

JOSEPH H. PRATT.

Harvard University Medical School, Boston, Mass., to whom all books and papers on these subjects should be sent for review.

Lewy. Die Beziehungen der Charcot-Leydenn'schen Krystalle zu den eosinophilen Zellen. Zeitschr. f. klin. Med. **40**: 59, 1900.

Lewy shows that where eosinophilic cells abound, there Charcot-Leyden crystals appear. This association is found in all the tissues in leukæmia, in the sputum in various diseases of the respiratory organs, in nasal polypi, in tumors, in the fæces in helminthiasis, and in the normal bone marrow. When the crystals are not present in the fresh preparations, they form quickly if the blood, pus, bits of tissue, or other material are preserved and kept from drying. The crystals can also be produced by the action of various metallic salts upon the eosinophilic cells.

The crystals can arise within the eosinphilic cells, and those that form outside the cells probably take their origin from eosinphilic granules, which lie free in the tissue-spaces. It is not to be assumed that the eosinophilic granules are directly transformed into crystals or simply supply the material by a chemical change. Lewy advances the hypothesis that the mother-substance of the crystals is formed physiologically in different tissues and is destroyed by the round cells attracted thither by chemotaxis. Under certain conditions the mother-substance is formed in such a large amount that a residue remains from which the crystals arise. The eosinophilic granules are formed from this mother-substance by the action of the round cells, which become transformed into eosinophiles. Lewy himself, however, brings forward objections to this hypothesis.

J. H. P.

Glińśki, L. K. Zur Kenntniss des Nebenpankreas und verwandter Zustände. Virchow's Archiv, **164**: 132–145, 1901.

A firm, oval, reddish gray, sharply circumscribed body, 4.5 cm. long, was discovered in the wall of the stomach near the pyloric end. It produced a bulging of the overlying mucous membrane into the cavity of the stomach. On microscopical examination the structure was recognized as an accessory pancreas. It was embedded in the muscular coat. In all the other recorded cases the accessory pancreas has been situated in the submucosa or between the serosa and the muscular layer. The author collected from the literature thirteen cases in which an accessory pancreas has been found. Three times it was located in the stomach wall; ten times in the wall of the intestine. The pancreas in some of the lower vertebrates is situated in the wall of the stomach or intestine, hence the accessory pancreas in man is regarded as a reversion to the original type.

J. H. P.

Warthin, A. S. A Contribution to the Normal Histology and Pathology of the Hæmolymph Glands. Journal of the Boston Society of Medical Sciences, **5**: 416–436, 1901.

Hæmolymph nodes differ from ordinary lymph nodes in that they contain blood-sinuses in place of the lymph-sinuses. These bodies were discovered by Gibbes in 1889. Six years later they were described in more detail by Robert-

son, who gave them the name of hæmolymph glands. Clarkson and others have studied the occurrence and minute anatomy of these organs in the lower animals, but little attention has been paid to the hæmolymph nodes of man. The author bases his report on the investigation of these structures in autopsies on eighty subjects. Hæmolymph nodes occur in greatest number in the prevertebral retroperitoneal region near the great vessels, near the adrenal and renal vessels, along the brim of the pelvis, and in the root of the mesentery. The differ as to location, number, and size in different individuals. They undergo atrophy in old age. They usually lie embedded in fat, and as a rule very near to the wall of some large vessel. An interesting and suggestive feature is the richness of their blood supply. The hæmolymph nodes cannot be definitely distinguished from the ordinary lymph nodes by naked-eye examination. This is owing to the fact that the blood sinuses are usually empty and collapsed after death. When the sinuses are filled with blood the bodies are deep red or bluish, and the smaller ones are easily mistaken for blood clots.

Two types of hæmolymph nodes exist, to which Warthin has given the names splenolymph gland and marrowlymph gland, as indicating their structure and probable function. Between these types are transition forms, and also between these bodies and the spleen on the one hand and ordinary lymph nodes on the other.

The splenolymph node is the more frequent form. It possesses a relatively thick capsule. Trabeculæ pass from this into the organ dividing it into irregular lobules. Branches of a peripheral blood sinus accompany the trabeculæ, increasing in size as they approach the center. Between the sinuses lies the lymphoid tissue. Round collections of lymphoid cells, suggesting splenic follicles, are common. Next to the small lymphocyte the large mononuclear cell is the most common form in the lymphoid tissue. Red blood corpuscles lie free in the meshes of the reticulum, and there is a varying amount of blood pigment. Mononuclear phagcoytes containing red blood corpuscles and blood pigment are found in the recticulum and in the central blood sinuses. Scattered areas of a hyaline substance which stains blue with Mallory's connective tissue stain occur in the lymphoid tissue. Fuchsinophile bodies, probably the product of the destruction of the red blood corpuscles, are seen in the reticular meshes and also in the mononuclear phagocytes. In the marrowlymph node there is a greater variety of cells than in the splenolymph node. Mononuclear eosinophiles are more numerous, and multinuclear cells and large mononuclear forms with deeply staining knobbed nuclei occur.

Warthin believes that under normal conditions the hæmolymph nodes are probably concerned chiefly in hæmolysis and leucocyte formation and play but little part, if any, in the production of red blood corpuscles. Under pathological conditions of the blood these bodies may assume a blood-forming function. The hæmolymph nodes take on the structure of either spleen or bone-marrow and compensate for these organs when their functional power is diminished by disease. J. H. P.

GENERAL PHYSIOLOGY.

RAYMOND PEARL.

Books and papers for review should be sent to Raymond Pearl, Zoölogical Laboratory, University of Michigan, Ann Arbor, Mich.

Schultze, L. S. Untersuchungen über den Herzschlag der Salpen. Jenaische Zeitschr. f. Naturwiss. xxxv, N. F. xxviii, pp. 221–328. Taf. ix–xi, 1901.

The alternation in the direction of the heart beat in the tunicates is a phenomenon which has frequently been observed and described, but the present paper is by far the most comprehensive and detailed study of the subject which has ever appeared. The work was done on several different species of the transparent pelagic tunicate *Salpa*. A complete period in the activity of the normal heart of *Salpa* includes a succession of series of advisceral and abvisceral pulsations, with an intervening short pause after each series. The different phases of the activity of the heart vary so widely, both absolutely and relatively, that it is impossible to give a normal value for any one of them. As an example illustrative of this great variability may be taken the relative number of pulsations in the advisceral and abvisceral series in the case of a specimen of *Salpa democratica–mucronata*. The advisceral pulsations were to the abvisceral as 100 is to 115 in one individual of the colony, while in another individual of the same size in the same colony the two series were related as 100 is to 45. The rate of the abvisceral and advisceral pulsations is in general the same. The condition of the water has a very decided influence on the activity of the heart. Stale water produces an increase in the number of pulsations and an acceleration in their rate. The author gives a detailed account of the phenomena observed in an animal slowly dying in foul water. The most significant appearance under these conditions is the loss of coördination in the heart beat. For example, abvisceral and advisceral pulsations may start from opposite ends of the heart at the same time and meet in the middle, neutralizing each other and disappearing. An advisceral series may be extended to a great length; in some cases to as many as 241 single pulsations without any pause. A section on the effects of poisons is mainly devoted to an account of the action of two drugs, nicotine and hellebore. Nicotine decreases the number of advisceral pulsations, while hellebore increases it.

Experiments were performed to discover the source of the cardiac stimulus. It was found that a heart completely isolated from the body beats in the normal manner, thus showing that the cause of the pulsation is not peripheral. To test the effect of the central nervous system on the heart beat, stimulation and extirpation experiments were performed. Electrical stimulation of the ganglion had no effect either on the number or the rate of the pulsations. Extirpation of the ganglion causes a decrease in the number of pulsations in a series, but it is shown that this is not a specific effect of the removal of the nervous system, but, instead, is a result of the loss of a certain amount of body substance. Experiments in which the heart was cut transversely in pieces gave the result that these pieces will, after a time, begin to beat rhythmically whether they are from the ends or the middle region of the heart. Emptying the heart of all blood did not affect the

normal beat. The motor stimulus which causes the rhythmical pulsation must arise in the muscles of the heart itself, since a very careful search with a great variety of histological methods failed to reveal either ganglion cells or nerve fibers in this organ.

The next general subject considered, is the cause of the periodical change in the direction of the blood flow and the heart beat. After a critical review of the theories which have been advanced by previous workers, Schultze proceeds to an account of his own explanation. By isolating one end of the heart he found that its activity showed a marked periodicity. There were periods of maximal activity, in which the pulsations were strong and rapid, followed by periods of minmal activity during which the beats were weakened and nearly disappeared. Both ends of the heart, under normal circumstances, would, of course, show this periodicity. From the fact that any single muscle fiber of the heart cannot be made by extra stimulation to further react when already contracted or nearly contracted, together with the fact that the stimulus is conducted in the muscle fibers themselves, it is shown that the end of the heart which is beating more rapidly and strongly will determine the beat of the whole. Taking this in connection with the periodicity in the activity of either end of the heart, the result is that first one and then the other end will determine the beat of the whole heart. When, for example, the abvisceral end is in its period of maximal activity it will set the whole organ to beating synchronously with it, outweighing and obscuring the weaker pulsations of the advisceral end. After a time, however, its period of maximal activity ends and that of the opposite end begins and controls in turn the beat of the whole. The continuation of this process results in the observed alternation in the direction of the heart beat and blood flow.

R. P.

Rádl, Em. Ueber den Phototropism einiger Arthropoden. Biol. Centralbl. **21**: 75–86, 1901.

This paper gives an account of the effect of light on the movements of the eyes of various Cladocera, and the relation of these eye movements to the phototaxis of the organisms. It was found that sudden shading of a Daphnia caused an immediate drawing in of the eye stalks. Careful study showed that under all conditions the eye was directed towards the source of greatest illumination. If, for example, a Daphnia on a slide on the microscope stage be shaded by the hand from above, the eye stalk will be rotated so as to point its tip towards the opening in the diaphragm; while, on the other hand, if the light be diminished from below, the eye will turn up towards the light now coming from above. When the organism (Daphnia) is oriented with its back towards the light the eye is in its normal position, with all the muscles of the stalk in a state of equal tension. If now the preparation is turned so as to bring the animal out of its position of orientation, it is found that the eyes maintain their orientation while the body turns about them as a fixed point until it is again in a position such that the eye muscles are in a state of equal tension. These reactions of the eyes do not usually appear in strong, direct sunlight, there being apparently an upper limit of intensity beyond which the normal phenomena do not appear.

Observations were made on the method of orientation to light of specimens

of *Simocephalus* swimming freely in the water. They always keep the back towards the source of light even though this necessitates an entire reversal of the usual position with reference to the force of gravity. Furthermore, to a sudden shading the animals react by a strong spring towards one side or the other. This results in getting all the individuals out of a shaded area in a short time.

The author considers the eyes of the Cladocera as physiologically comparable with the statocyst of the decapod crustacea. The eye orients itself along the "lines of force" of the light rays, and thus effects differences of muscle tonus. On the other hand, the statolith moves along lines of the force of gravity to the lowest point of the statocyst and, through the sense hairs, causes differences in the tonus of the body muscles. The general biological significance of the phototactic reaction is discussed, and the orientation of swarms of *Culicidæ* to surrounding objects is explained as such a reaction. R. P.

CURRENT BACTERIOLOGICAL LITERATURE.

H. W. CONN.

Separates of papers and books on bacteriology should be sent for review to H. W. Conn, Wesleyan University, Middletown, Conn.

Karlinski. Zur Kenntnis der säurefesten Bakterien. Cent. f. Bak. u. Par. 1, **29**: 521, 1901.

Murray. A preliminary report on acid resisting bacilli, with special reference to their occurrences in the lower animals. Jour. of Exp. Med., p. 205, 1900.

Karlinski has made a study of the nasal cavities of quite a large number of individuals, originally for the purpose of determining whether the lepra bacillus could be found in these cavities in people not suffering from leprosy. In the course of this study he has discovered, in the nose in 19 cases out of 235, a very characteristic bacillus, which holds its stains against the action of acids in the same manner as the tuberculosis bacillus. This bacillus is larger than the tubercle bacillus or the lepra bacillus, and, indeed, when compared with the various other "säuerfest" bacilli, proved to be quite different from any of them. It appears to be the nearest to the organism discovered by Rabinovitsch, although, in some respects, it is different from that variety. The author thinks it is a new type of bacillus holding stains against the action of acids. The organism does not appear to be pathogenic for animals or for man when simply placed in the nasal cavities, although it is commonly found in individuals showing certain ulcers in the nose.

The second author studies the bacilli from the genital organs of dogs, horses, cows, cats, guinea pigs, rabbits, and white rats. He finds in all cases, except in those of cats and rabbits, acid resisting bacilli, resembling the smegma bacillus. They are not all alike, and the author thinks they form a group of closely allied but variable bacteria. H. W. C.

Reichenbach. Ueber Verzweigung bei Spirillen. Cent. f. Bak. u. Par. 1, **29**: 553 1901.

During recent years many questions have been raised in regard to the relations of bacteria to other fungi, and there are many who have a strong suspicion, amounting to a belief, that they are

to be regarded as stages in the development of the higher fungi. This conclusion is no new one, inasmuch as it was advanced in the early days of bacteriological study; but it has been revived in recent years, because of evidence based upon quite new data. The most important fact pointing in this direction has been the discovery among some bacteria, for example the tubercle bacillus and the diphtheria bacillus, of undoubted branching forms. Branching is not supposed to be characteristic of typical bacilli, and wherever it occurs has suggested a relation to some of the higher fungi. Our author believes that the evidence for the branching of bacilli hitherto advanced is not quite conclusive, being of the opinion that many or most of the facts may possibly be explained upon the ground that the branching forms are degenerate types. He conceives that the spirilli are the most promising organisms for the proper solution of the question, and makes, therefore, a careful study of *Spirillum rubrum*. Under proper culture media he is able to obtain undoubted instances of branching forms of this spirillum, several excellent photographs of which are given. Whether these branching forms are to be regarded as normal or as degenerate types, he is unable positively to ascertain, inasmuch as the various branches do not all show the typical spiral coiling, and he concludes that if the branching is a normal feature, every branch should become spirally coiled and should, perhaps, subsequently show branching in turn. These characters he does not find, and while, therefore, he is confident that these spirilli have a true branching, he is unable to determine positively whether it can be regarded as a normal or abnormal character. The question of the relation of bacilli to the higher moulds is therefore left still uncertain.

H. W. C.

Bliesener. Beitrag zur Lehre von Sporenbildung. Zeit. f. Hyg. **32**: 71, 1901.

Some investigators of cholera epidemics have reached the conclusion, from various facts connected with the distribution of the disease, such as the breaking out of the disease anew in the spring after a season of winter weather, or its sudden appearance in localities after having disappeared for a long time, that this bacillus must, under certain circumstances, produce spores or, at least, resisting forms capable of lying dormant for a long time. The author has obtained direct evidence that something of this kind occurs. A quantity of water contaminated with a large amount of organic material was filtered, and subsequently sterilized by discontinuous heat; into this a small amount of cholera bacillus was inoculated and, after some time, the author finds in the precipitate, which appears at the bottom, a number of oval, highly refractive bodies, which, in appearance and their relation to stains resemble spores. Experimental evidence which followed showed him that these were forms of the cholera bacillus, since they develop into the typical forms. He is inclined to believe, therefore, that they are spores of the cholera bacillus, but recognizes that the conclusion is not very satisfactory, inasmuch as a half hour heating at a temperature of 50° C is sufficient to destroy the vitality of these bodies, whereas true spores resist a much higher heat. In spite of this, the author is inclined to believe that he has discovered a spore formation in the cholera bacillus.

H. W. C.

NOTES ON RECENT MINERALOGICAL LITERATURE.

ALFRED J. MOSES AND LEA MCI. LUQUER.

Books and reprints for review should be sent to Alfred J. Moses, Columbia University, New York, N. Y.

Clark, F. W., and Steiger, G. The Action of Ammonium Chloride upon Natrolite, Scolecite, Prehnite, and Pectolite. Am. Jour. Sci. iv, **9**: 345, 1900.

The data given yield further proof of the *availability* of the method for investigation into the chemical constitution of silicates.

Natrolite and *Scolecite* proved to have no constitutional water, but similar chemical formulæ, which, however, are not those of ortho-silicates:

Natrolite, $Na_2Al_2Si_3O_{10} \cdot 2H_2O$.

Scolecite, $Ca\ Al_2Si_3O_{10} \cdot 3H_2O$.

Prehnite may be regarded as having constitutional water and ortho-silicate formula.

Pectolite differs widely from these other minerals as regards the ammonium chloride reaction; but experiments suggest the probable existence of a hydrous ammonium pectolite.

L. McI. L.

Clark, F. W. and Steiger, G. The Action of Ammonium Chloride upon Analcite and Leucite. Am. Jour. Sci. iv, **9**: 117, 1900.

Authors show that both analcite and leucite, when heated to 350° with this reagent in a sealed tube, yielded the same definite compound, ammonium leucite $NH_4Al\ Si_2O_6$. This reaction was fairly general in character, and analagous results were obtained with other species.

By substituting in many minerals a volatile base for fixed bases, silicates are obtained which split upon ignition in such a way as to shed light upon their molecular constitution.

The conclusion was reached that analcite and leucite were not true metasilicates, but pseudo mixtures.

L. McI. L.

Parsons, Charles L. The Use of Metallic Sodium in Blowpipe Analysis. Jour. Am. Chem. Soc. **23**: 159, Mar. 1901.

Contrary to the statement of Hempel, who first proposed the use of sodium in blowpipe analysis, the author finds that the reduction of metallic compounds by means of sodium takes place with the greatest ease on charcoal. The following procedure is recommended: A piece of metallic sodium 3 or 4 mm. in diameter is hammered flat. The powdered substance to be tested is spread upon and pressed into the metal, and the whole turned into a little ball. It is placed in a slight depression of a piece of charcoal, and ignited with a match or Bunsen flame. The residue is heated before the blowpipe, and fusible metallic particles readily collect into a globule; at the same time coatings of the volatile metals are obtained. Treated in this way, garnierite gives magnetic nickel, chrysocolla a copper button, and cassiterite a tin

button, as readily as a lead button is obtained from cerussite. Large quantities of sodium are to be avoided, and care must be taken not to touch the metal with the fingers, as the reaction sometimes begins spontaneously. A. F. R.

Morgan, Leonard P., and Smith, E. F. Experiments on Chalcopyrite. Jour. Amer. Chem. Soc. **22**: 107, Feb. 1901.

Weighed portions of chalcopyrite were exposed to the action of dry hydrochloric acid gas in a heated combustion tube. The liquid in the tube was titrated with potassium permanganate, and gave an iron content of from 30.56 to 30.72 per cent., theory requiring for chalcopyrite 30.5 per cent. Results indicate complete decomposition, and show that all the iron is in a ferrous state. The results obtained by heating the mineral in a closed tube with a solution of copper sulphate confirm this.

A. F. R.

Atkinson, Elizabeth Allen. Indium in Tungsten Minerals. Jour. Amer. Chem. Soc. **20**: 811, 1898.

From 150 to 300 grams of wolframite, hübnerite, and scheelite, from several localities, were carefully examined for indium, but only in the wolframite from Zinnwald was any found. Author comes to the conclusion that indium cannot be regarded as an associate of tungsten, and that Hoppe-Seyler's suspicion as to blende being its origin is probably correct, for only in the Zinnwald mineral was it found, zinc also being present only there. A. F. R.

Vater, Heinrich. Ueber den Einfluss der Lösungenossen auf die Krystallisation des Calciumcarbonates. Theil viii, Zeit. f. Kryst., **31**: 538–578, 1899.

This paper discusses the action of the solution upon gypsum and anhydrite, and concludes that at lower temperatures, below 30°C., calcite alone results, and that the only known cause by which pure calcium carbonate separates as aragonite is a temperature exceeding 30°C. A. J. M.

Goldschmidt, V. Ueber Trögerit und künstlechen Uranospinit. Zeit. f. Kryst. **31**: 469–478, 1899.

Concludes trögerite is tetragonal, not monoclinic, and ventures supposition that all other uranium micas: autunite, uranospinite, torbernite, zeunerite, and phosphuranylite are also tetragonal.

A. J. M.

Viola, C. Zur Kentniss des Anorthits vom Vesuv. Zeit. f. Kryst. **31**: 484–498, 1899.

Crystallographic study.

Ward, H. L. Notice of an Aerolite that recently fell at Allegan, Mich. Am. Jour. Sci. iv. **8**: 413, 1899.

The stone is light ash-gray in color, very friable, and covered with a black crust, which is 1–2 mm. thick, and has a smooth or wavy surface. The structure is chondritic, and the following minerals are present: enstatite, chrysolite, feldspar, troilite and iron, the two latter being granular and evidently scattered. G.=3.558. L. McI. L.

Grimsley, G. P., and Bailey, E. H. S. Report on Gypsum and Gypsum Cement. Vol. v, Univers. Geol. Sur. of Kansas.

MEDICAL NOTES.

METHODS FOR STAINING TUBERCLE BACILLI.

EHRLICH–WEIGERT ANILIN–METHYL–VIOLET METHOD. — Place a dried cover-glass preparation, film down, in the following solution, and heat gently until steam rises, then allow to stand for 2 to 5 minutes:

Methyl violet, sat. alc. sol.,	1.1 part.
Alcohol, absol.,	1. part.
Anilin water,	10. parts.

Anilin water is made by using 1 part anilin oil with 20 parts of distilled water, and after standing a short time and becoming thoroughly saturated, filtering the mixture. Decolorize for a few seconds in 1 part nitric acid and 3 parts water. Wash one or two seconds in 60 per cent. alcohol, then in water. If it is desired to counterstain the specimen, allow a few drops of saturated aqueous solution of vesuvin to cover the specimen for about five minutes. When the staining is complete the preparation is washed, dried, and mounted in balsam.

GABBETT'S METHOD.—This method is simple and rapid, and is considered by many to be a most excellent method for routine work. The cover-glass preparation is stained for 2 to 5 minutes in Ziehl's carbol fuchsin solution, after the formula:

Fuchsin,	1 part.
Alcohol,	10 parts.
Carbolic acid (5 per cent. sol.),	100 parts.

The fuchsin should be dissolved in the alcohol before the acid is added. After this solution is allowed to act for the desired length of time, the preparation is placed for 1 to 2 minutes in Babbett's methylen-blue sulphuric acid solution, consisting of:

Methylen blue,	1 part.
Sulphuric acid (25 per cent. sol.),	50 parts.

The specimen is then washed in water, dried, and mounted in balsam. Tubercle bacilli are stained red, while other elements of the mount are blue.

ZIEHL–NEELSON METHOD.—By this method tubercle bacilli are stained with the following solution:

Fuchsin, 1 part, dissolved in 10 parts alcohol, to which is added 100 parts 5 per cent. solution of carbolic acid. The cover-glass preparation is floated, film down, on the solution, to which gentle heat is applied until steam rises. The specimen is then washed in water, and decolorized in 25 per cent. nitric or sulphuric acid, then in 60 per cent. alcohol for a very short time, after which, with thorough washing in water, it is mounted in balsam. C. W. J.

NEWS AND NOTES.

The New Haven Microscopical Society has just issued a very neat booklet containing the constitution and by-laws of the society, as well as a list of the members with the address of each. The officers of the society are: President, Robert Brown, Observatory place, New Haven; Secretary and Treasurer, J. F. Malone, 25 Wooster place, New Haven.

The annual meeting of the American Microscopical Society will be held in Denver, Col., August 29 to 31. Efforts are being made to secure exceptionally low rates, and an enthusiastic and profitable meeting is assured.

The Hopkins Seaside Laboratory, of Leland Stanford Junior University, began its tenth session at Pacific Grove on Monterey Bay, Monday, June 10, 1901. The regular courses of instruction continue six weeks, closing July 20th. Provision is made at the laboratory to accommodate three classes of students: (1) teachers and students who wish to pursue laboratory courses in botany and zoölogy; (2) advanced students in zoölogy, physiology, and botany; and (3) investigators who are prepared to carry on researches in morphology and physiology. The regular courses, with the instructors in charge, are as follows:

1. General Zoölogy—Professor G. C. Price, Leland Stanford Jr. Univ.
2. Elementary Botany—Professor S. C. Price.
3. Advanced course on Structure and Physiology of Algæ.—Professor G. J. Peirce, Leland Stanford Jr. Univ.
4. Embryology—Professor G. J. Peirce.
5. Comparative Morphology and Histology of the Nervous System and Sense Organs.—Professor F. M. MacFarland, Leland Stanford Junior University.
6. Advanced course in Zoölogy.—Professor F. M. MacFarland.
7. General Ornithology.

We have received the announcement of the third session of the Rhode Island Summer School for nature-study to be held at the Rhode Island College of Agriculture and Mechanic Arts, Kingston, R. I., July 5 to 20, 1901. The instruction is to be almost wholly in the nature of field work, the schedule being made up of excursions, led by competent men in every branch of natural science. The time given to class-room exercises will be just enough to indicate what is to be observed and done in the field. Special evening lectures will be given. Communications should be addressed to "Summer School," Kingston, R. I.

QUESTION BOX.

Inquiries will be printed in this department from any inquirer.
The replies will appear as received.

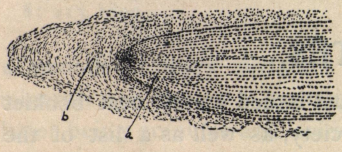
8. Where can "Stabilite" insulating material be bought? Is it used much in American laboratories? L. H.

9. What is the best method of drying and mounting microscopic fungi? Can you refer me to a good book dealing with the subject. M. R.

REPLIES TO QUESTIONS.

"What is meant by the *growing* tip in allium?" (Question No. 1.)

The growing part of a root (*a*), or the "growing tip," is a short space about one-sixth of an inch back from the extreme end. In this part of the root the cells divide rapidly, and its length is thereby increased. This is the only part of the root in which growth takes place. The end of the root is usually covered by a protecting coat of dead cells (*b*), derived from the living zone just back of it. These dead cells constitute what is known as the root cap.



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"My sister was troubled with nervous headaches while she used coffee. She found how greatly improved we were from discontinuing it and using Postum Food Coffee, so she made the change, and is now rid of her nervous headaches. We are naturally strong advocates of Postum."—Mrs. J. Walford, Castalia, Erie Co., Ohio.

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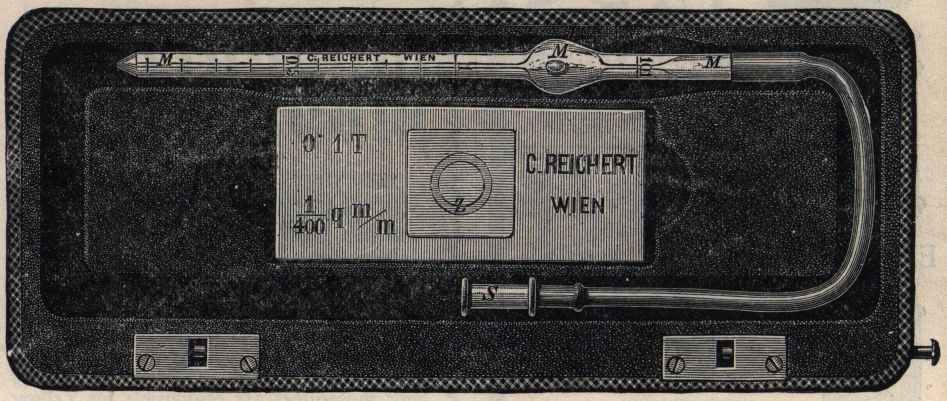
Let one meal in the day consist of an abundance of good meat, potato and one other vegetable.

This method will quickly prove the value of the selection of the right kind of food to rebuild the body and replace the lost tissue which is destroyed every day and must be made up, or disease of some sort enters in. This is an age of specialists, and the above suggestions are given by a specialist in food values, dietetics and hygiene.

Original Articles in the December Number, 1900.

ARTICLES IN JAPANESE.

HAEMACYTOMETER AFTER THOMA.



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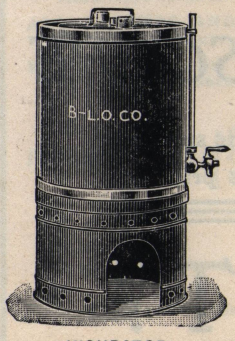
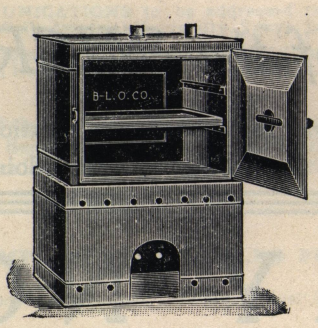


INCUBATOR.



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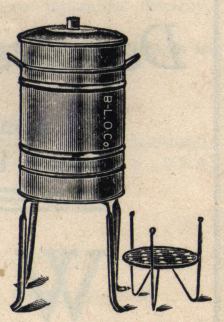

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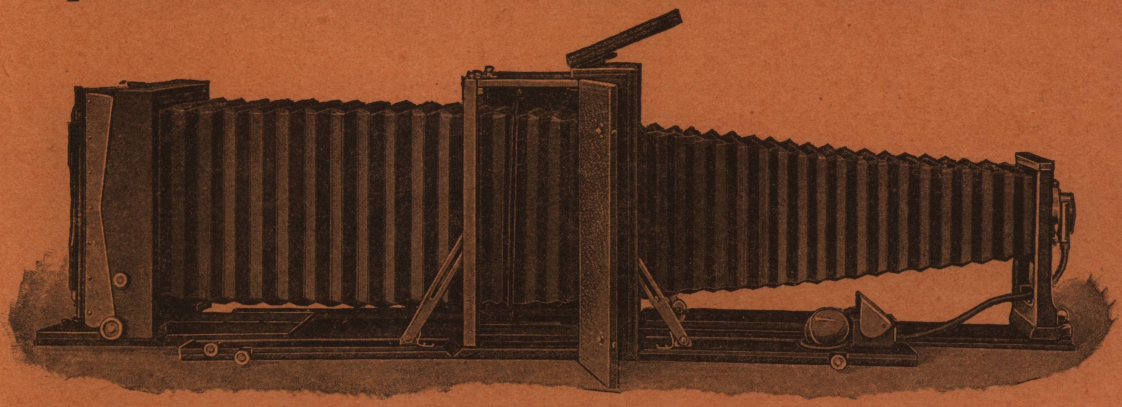

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